Advances in Equine Health and Management II: Grazing Winterhardy Perennial Ryegrass, Estimating Bodyweight, and Evaluating Novel Methodologies for Livestock Water Quality

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Abstract

The horse industry provides $122 billion dollars of value added to the U.S. economy, with a $50 billion dollar direct impact to GDP (AHC, 2017a). The use of horses is incredibly diverse, ranging from rescues and recreation sectors to working horses and the competition and racing sectors. Horse owners and enthusiasts are a welcoming community with a strong desire to educate themselves and improve the health of their horses, increase production of their land, and minimize labor and feed costs. In the age of digital media, there are numerous online forums and popular press websites to share information. In addition to, and perhaps despite this, many owners look for scientific information to validate their management decisions. This feedback allows us to keep track of questions and issues that horse owners are having today. In turn, this leads us to new research to answer these questions. Three specific areas of interest are highlighted in this dissertation—alternate forages and ways to improve existing pastures, how to manage horse bodyweight and body condition, and ways to ease or reduce the labor involved in farm management.

Perennial ryegrass (PRG) is a high quality and yielding forage that, until recently, lacked the winterhardiness to persist in the upper Midwest. The objective was to evaluate the yield, persistence, forage nutritive value, and livestock preference of a new winterhardy PRG cultivar ('Forageur') grown in monoculture and in mixture with white clover under horse and cattle grazing. Research was conducted in St. Paul (SP) and Grand Rapids, MN (GR). Meadow fescue (MF), tall fescue (TF), orchardgrass (OG), and three perennial ryegrass varieties ('Forageur', 'Remington', and 'Spreader IV') were

seeded in 2015 (SP) and in 2016 (GR); and were grazed for 2 days by either 6 adult horses (SP) or by 6 cow-calf pairs (GR). Prior to grazing, forage yield and nutritive value samples were collected, and post-grazing, fields were visually assessed for the percentage of forage removal to determine livestock preference. Yields of PRG cultivars were similar but lower yielding (6.9 to 7.6 Mg ha^{-1}) compared to other CSG. Persistence was similar among all CSG in SP; however it was lower for PRG in GR. 'Forageur' was highly preferred by both horses (≥74% removal) and cows (≥70% removal) and was consistently among the highest for crude protein (CP; 200 g kg^{-1}), equine digestible energy (Equine DE; 2.35 Mcal/kg), and bovine metabolizable energy (Bovine ME; 2.6 Mcal/kg), moderate for nonstructural carbohydrates (NSC; 136 g kg^{-1}), and among the lowest for neutral detergent fiber (NDF; 518 g kg^{-1}). This research suggests that 'Forageur' is viable PRG cultivar to use in upper Midwest livestock pastures.

Adding breed type, height, and neck circumference to body length and girth circumference improves bodyweight (BW) estimation in different breeds of horses; however, equations have not been developed for all breed types. The objectives were to develop BW estimation equations for Miniature, saddle-type, and Thoroughbred horses using morphometric measurements. Measurements were collected on adult (≥ 3 yr, non-pregnant) saddle-type (n=209), adult (n=249) and juvenile (< 3 yr, n=61) Miniatures, and adult Thoroughbreds (n=100). Personnel determined body condition score (BCS), measured withers height and girth circumference at the third thoracic vertebra, body length from the point of the shoulder to the point of the buttock and to a line perpendicular to the point of the buttock, and neck circumference at the midway point between the poll and withers. Each horse was weighed using a livestock scale. Bodyweight estimations equations were developed using linear regression modeling and log transformation. Mean (±SD) BCS was 6.1 (±0.8), 5.4 (±0.6), 6.0 (±1.0) and 5.0 (±0.6) for adult and juvenile Miniatures, saddle-type, and Thoroughbreds, respectively. Bodyweight estimation equations developed through the current research were within 4% of the scale BW, and offered improvements over previous BW estimation equations and weight tapes, which were inaccurate by 5 to 25%. Owner estimated BW was within 8 to 15% of scale BW. Morphometric measurements were successfully used to develop BW equations for Miniature, saddle-type and Thoroughbred horses. The equations will assist owners and professionals with managing horse BW and will be added to the Healthy Horse app.

Goldfish (*Carassius auratus*) have been reported as a method to keep water tanks clean; however, little information exists on this approach. The objectives were to evaluate the efficacy of goldfish on maintaining water quality in tanks and to evaluate the frequency that this method is used. The first objective was completed June through October 2017 in St. Paul, MN using plastic and metal 379 L stock tanks, each with and without goldfish in a drylot that housed six adult horses. The stocking rate was 5 goldfish per tank, and all tanks were refilled when any tank reached 190 L. Daily readings of total dissolved solids (TDS) and water turbidity (NTU), and weekly samples to measure chlorophyll *a* were taken. At the end of each 28-day period, tanks were cleaned and rotated. Plastic tanks had lower TDS compared to metal tanks ($P < 0.001$); however, metal tanks had lower NTU and chlorophyll *a* ($P \leq 0.008$). Adding goldfish resulted in

lower TDS ($P < 0.001$); however, there was no effect on NTU or chlorophyll *a* ($P \geq 0.097$). The second objective was completed using an online survey that was open from October 31 until December 15, 2018. Of the 672 completed surveys, 56% had not tried using goldfish in water tanks, 26% had utilized goldfish in the past, and 18% currently used goldfish. The inclusion of goldfish in water tanks did not affect all water quality parameters; however, 44% of survey respondents had tried, or were currently using, this management method.

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CHAPTER ONE: LITERATURE REVIEW

Introduction

The economic impact of the horse industry is tremendous. The horse industry provides $122 billion dollars of value to the US economy, with a $50 billion dollar direct impact to GDP (AHC, 2017a). There are 988,394 jobs directly tied to the horse industry with an overall employment impact of 1,744,747 jobs (AHC, 2017a). There are 7.2 million horses in the United States and 30.5% of all US households contain at least one horse enthusiast (AHC, 2017a). Overall, 80.9 million acres of land in the US are used for horse related purposes (AHC, 2017a), and in Minnesota (MN), 145,727 horses are kept or used on 360,000 acres of land (AHC, 2017b). Horses add $1 billion dollars of value to the MN economy and provide 21,039 jobs (AHC, 2017b).

Unlike many other livestock species, the use of horses is incredibly diverse, ranging from rescues and recreation sectors to working horses and the competition and racing sectors. Although there may be more of a spotlight on elite competition and race horses, the recreation sector is an area that deserves as much attention, particularly in Minnesota. Specifically in MN, the recreation sector supports 7,181 jobs and adds $341 million dollars to the economy (AHC, 2017b). Although the dollar value of the recreation sector is lower than the competition sector within MN ($652 million), the job impact is similar (7,666 jobs; AHC, 2017b). This emphasizes the need to support and provide resources for small acreage horse owners and larger acreage boarding facilities equally.

Horse owners and enthusiasts are a welcoming community with a strong desire to educate themselves and improve the health of their horses, increase production of their land, and minimize labor and feed costs . In the age of digital media, there are numerous online forums and popular press websites to share information. In addition to, and perhaps despite this, many owners look for scientific information to validate their management decisions. The Extension branch of the University system was created to facilitate the translation of research data to the everyday information consumer. Specifically at the University of Minnesota, the equine Extension program reaches nearly 3 million people annually using Facebook, YouTube, an e-newsletter, website, and in-person events. Not only are we able to share the latest data and information from our research, as well as from other universities, we also receive valuable information via consumer feedback. This feedback allows us to keep track of questions and issues that horse owners have. In turn, this leads us to new research questions, including alternate forages and ways to improve existing pastures, how to manage horse weight and body condition, and ways to ease or reduce the labor involved in farm management.

Horse Forage Requirements

Horses, as well as other livestock including cattle, are herbivores, meaning they meet their daily nutrient requirements by consuming plant matter. The unique aspect of the horse compared to other livestock, such as cattle (ruminants) and other monogastrics such as swine, is the fermentation that occurs in the hindgut. As hindgut fermenters with a relatively small stomach, horses are designed to eat forage continuously throughout the day (Janis, 1976). Forages are characterized by a high fiber content, which consists of both structural carbohydrates (SC) and nonfiber carbohydrates (NRC, 2007). Microbial fermentation of the SC in the cecum of the large intestine produces volatile fatty acids, which are thought to provide 60 to 70% of the horse's total energy needs (Vermorel and Martin-Rosset, 1997). Voluntary dry matter intake for grazing horses generally ranges from 1.5 to 3.1% of bodyweight (BW; NRC, 2007). It is recommended that forages make up at least 50% of an adult horse's diet (NRC, 2007; Becvarova et al., 2009).

Research has demonstrated that a diet high in forage content, whether as hay or fresh pasture, helps to reduce negative behaviors including coprophagy (eating of feces), bedding-eating, cribbing, and undesirable chewing (Pell and Mcgreevy, 1999; Hothersall and Nicol, 2009). Hay is a dried forage, and the primary benefit of it is that it can be stored and fed year round. However, it has been observed that horses fed hay and grain spend much less time chewing compared to horses on pasture, resulting in less saliva present to moisten the food. This can affect gut acidity, and may lead to dysfermentation and colic, as well as ulceration in the hindgut (Harris et al., 2006; Ralston, 2007). Decreased access to pasture also increases the risk factor for colic (Hudson et al., 2001).

In addition to considering management strategies from a horse health viewpoint, it is important to consider them from an economic standpoint. Feed costs are one of the greatest expenditures of horse ownership, and in particular, the cost of purchased and stored feed is high relative to the cost of feeding horses and other livestock from pasture (McCormick et al., 2006). Pastures have the capacity to meet or exceed the daily nutrient requirements for most classes of horses (NRC, 2007) and offer the advantage over hay in that they are a less labor intensive feeding system (McCann and Hoveland, 1991). Martinson et al. (2006) conducted a state-wide survey of MN horse owners and determined that 86% of horses are kept on pasture or grassy turnout during the summer. Results from the survey also highlighted the percentage of horse owners with lower income as 25% of respondents reported pre-tax incomes of less than $50,000. In addition to that, the topic of pasture management was one that respondents had the least amount of knowledge in. Therefore, maximizing pasture health and productivity can greatly reduce reducing feed costs, as well as overall costs associated with maintaining a pasture.

Pasture as a Forage Option

Grasses can be classified as either annual or perennial, as well as either warm-season or cool-season. Annuals only live through one growing season, and need to be re-seeded yearly. Perennials return yearly from the same root system, and thus do not need to be re-seeded as frequently. Warm-season grasses are those that survive and thrive in a tropical or sub-tropical climate, whereas cool-season grasses (CSG) survive and thrive in a more temperate environment. The primary reason for these differences is the photosynthetic pathways within each type of grass. Perennial grasses are an important component of pastures that provide year-round ground cover and offer an affordable form of forage for a diversity of livestock. Producers and horse owners tend to select forage species based on a number of factors including yield, persistence , preference, and nutritional content (Rojas-Downing et al., 2018).

Cool-Season Grasses

In the upper Midwest, perennial CSG are the foundation of productive pastures (Allen et al., 2012), as they are best suited for the local environment and offer the best cost-benefit balance for those looking to limit economic and labor inputs. Pasture species, and/or variety selection, should be based on yield and persistence, animal preference, and the nutritive value of the plant.

Yield. Research by Allen et al. (2012; 2013) and Martinson et al. (2016) evaluated perennial CSG under horse grazing. Orchardgrass (OG; *Dactylis glomerata* L.) had higher yields (10.1 to 13.9 T ha^{-1}) compared to other grasses including smooth bromegrass (*Bromus inermis* Leyss.), meadow bromegrass (*Bromus biebersteinii* Schult.), creeping foxtail (*Alopecurus arundinaceus* Pior.), Kentucky bluegrass (KBG; *Poa pratensis* L.), meadow fescue (MF; *Schedonorus pratensis* Huds.), perennial ryegrass (PRG; *Lolium perenne* L.), quackgrass (*Elymus repens* L.), reed canarygrass (*Phalaris arundinacea* L.), tall fescue (TF; *Schedonorus phoenix* Scop.), and timothy (*Phleum pretense* L.). Kentucky bluegrass, MF, PRG, and TF ranged from 9.4 to 11.1 T ha^{-1} in year one and from 7.3 to 9.6 T ha^{-1} in year two. Allen et al. (2012) remarked that year one of the study had an early spring (April) and wet fall, which allowed for seven months of grazing versus year two, which had a typical spring (May) and dry fall, resulting in five months of grazing. This resulted in an average difference of 2.4 T ha^{-1} between years. Weather, particularly in the springtime, plays a large role in dictating the grazing season in the upper Midwest. If the spring “green up” is delayed due to colder temperatures, horse owners will need to delay pasture opening past the typical start of the season (May). This can potentially place economic strain on owners that need to find additional hay for their horses past their planned winter allotment. Martinson et al. (2016) evaluated twelve different CSG mixtures, with yields ranging from 6.1 to 7.1 Mg ha^{-1}. Most seeding recommendations for pastures are based on a mixture of species, as this results in a diverse pasture that can withstand stress from the environment and from the animal.

Furthermore, Marten and Hovin (1980) reported yields ranging from 7.2 to 11.0 Mg ha^{-1} for OG and 5.4 to 7.0 Mg ha^{-1} for TF in a 4-cut system in Minnesota. Casler and van Santen (2001) reported means for TF and MF that ranged between 8.3 and 8.6 Mg ha^{-1} under cow and heifer grazing. Studies completed in both Mississippi (Solomon et al., 2014) and the United Kingdom (Gibb and Baker, 1989; Orr et al., 2005; Marley et al., 2014) reported PRG yields ranging from 1.3 to 3.9 Mg ha^{-1}. This range is considerably lower than PRG yields reported by Allen et al. (2012) despite similar or greater seeding rates. All studies utilized rotational grazing and/or careful management of post-grazing height, and the explanation for the relatively decreased yield is unclear. Yields for PRG under sheep and cattle grazing in New Zealand were greater than other published research, and is likely due to favorable weather conditions and a long growing season. Yields ranged from 10.9 to 14.2 t ha-1 for 21 PRG cultivars (Easton et al., 2001).

White clover (*Trifolium repens* L.) is often added to CSG pastures, especially PRG pastures, to reduce the dependency on nitrogen fertilization (Collins and Rhodes, 1989). PRG-white clover pastures under beef cow grazing yielded more (10.59 t ha^{-1}) compared to pastures harvested for silage (8.06 t ha^{-1}) or under mechanical harvest (7.18 t ha^{-1}; Kleen et al., 2010). Wilman and Asiegbu (1982) reported lower yields of PRG-white clover mixtures on a 4-week harvest schedule (3.5 to 4.1 Mg ha^{-1}). Similar PRG and PRG-white clover mixture yields were observed on a 28-35 day cycle for dairy cow grazing in France (4.7 to 5.3 mg ha^{-1}; Ribeiro Filho et al., 2005). Overall, herbage mass did not differ between grass and grass-clover swards; however, PRG swards were taller than PRG-clover swards. In 2019, Ribeiro Filho et al. (2019) conducted a similar experiment to the 2005 study, and reported similar yields for PRG (4.5 mg ha^{-1}) and PRG-white clover (3.3 mg ha^{-1}) under a 35-day rotation. Remarkably, the pastures used in the 2019 study were seeded in 1998, which demonstrates longevity of both species used (Ribeiro Filho et al., 2019). Schils et al. (1999) reported greater yields of PRG-white clover swards, ranging from 14.6 to 10.8 t ha-1, and noted that overall plots that were both rotationally grazed by dairy cows and cut had more herbage mass than plots that were only mechanically harvested. Elgersma et al. (1998) reported results consistent with others, with PRG-white clover swards yielding more (7.7 to 13.3 t ha^{-1}) than PRG swards (2.3 t ha^{-1}) under mechanical harvest.

Persistence. Persistence is a critical quality within forages. If a species does not persist well under livestock grazing, or within colder climates, the producer will need to spent time and money re-seeding a field. Persistence has been evaluated for common perennial CSG species under horse and cattle grazing, as well as for other forage species. Olson et al. (2011; 2016) has done extensive work evaluating the effects of close, continuous grazing by horses on OG, timothy, smooth bromegrass, KBG, PRG, and TF.

Overall, OG and TF tolerated overgrazing well, while timothy did not. Timothy is an erect species, sensitive to heavy defoliation, and known to not do well under horse grazing (Olson et al., 2011; Allen et al., 2012; Martinson et al., 2016). Kentucky bluegrasses and PRG varieties varied in tolerance to grazing. Martin and Hovin (1980) reported similar findings in that OG and TF persisted better in a three-cut system than bromegrass, and that both OG and TF persisted better in systems with more frequent defoliations. Although the grasses in the study were mechanically harvested, the data is still useful when considering practical applications for a rotational grazing system. Brummer and Moore (2000) evaluated perennial cool-season grasses under continuous grazing by beef cattle in Iowa. Grasses were not grazed until the first production year; however, PRG did not survive (9% persistence) the first winter and was eliminated from the study. In a comparison of commonly grown CSG species, PRG was moderately ranked for persistence after two years; however, it had a reduction in yield from the seeding year to the first production year of about 26% (Allen et al., 2012). Elgersma et al. (1998) evaluated PRG-white clover mixtures under mechanical harvest in the Netherlands, and reported both winterkill and winter injury in all plots when winter temperature and precipitation was below average. Although PRG is known to be sensitive to cold temperatures during the non-growing season, it has also been demonstrated to be sensitive to heat and low rainfall (Smith et al., 1998), with fields under natural rainfall having lower yield than irrigated fields. This is similar to the “summer slump” discussed by Grev et al. (2017) and DeBoer et al. (2017), in which CSG pastures typically lag in yield production in the warmer/drier summer months (July/August) compared to spring and fall months. Ground cover was evaluated for 21 PRG cultivars in New Zealand, and the authors reported greater percent ground cover for denser cultivars, compared to the cultivars that were either more erect or those that mature faster, as well as compared to the oldest cultivar in the trial (Easton et al., 2001).

Preference. Previous work has evaluated cool-season grass preference under both horse and cattle grazing. Allen et al. (2013) reported moderate to high preference towards PRG by horses grazing CSG (46 to 95% removal). Orchard grass was less preferred than RG, while TF and MF were similar to PRG (Allen et al., 2013). Allen et al. (2013) also reported observations of coarse leaf edges in TF, and hypothesized that this could have negatively affected horse selection. Archer (1978, 1980), Hunt and Hay (1990), and Olson et al. (2011) reported moderate to high preference of PRG compared to other cool-season grasses under horse grazing. Casler and van Santen (2001) found Holstein cows and heifers grazing MF and TF preferred MF. Solomon et al. (2014) reported cattle preference for tetraploid PRG cultivars over diploid PRG cultivars, which appeared to be linked with herbage mass. McCann and Hoveland (1991) also reported a negative correlation between preference and maturity, whereas Allen et al. (2013) did not find a significant correlation between the two parameters. Allen et al. (2013) found a positive correlation between NSC and preference; however, did not find a correlation between preference and CP or maturity. Reid et al. (1967), Longland and Byrd (2006), and Smit et al. (2006) also reported a positive relationships between preference and carbohydrates; however Catalano et al. (2019) reported a negative correlation between NSC and preference by horse grazing legumes. Previous research has also reported positive relationships between preference and equine DE (Catalano et al., 2019), CP content (Fontenot and Blaser, 1965; Catalano et al., 2019), and a negative relationship between preference and height (Fleurance et al., 2010; Catalano et al., 2019), maturity (Burton et al., 1956; McCann and Hoveland, 1991; Catalano et al., 2019), and fiber content (Fontenot and Blaser, 1965; Smit et al., 2006; Allen et al., 2013). Many factors can affect animal preference, including available species, agronomic management, geographic location, and weather conditions (Marten, 1978).

Forage Nutritive Value. Allen et al. (2013) reported cool-season CP values that ranged from 167 to 253 g kg^{-1}, with PRG values ranging from 202 to 253 g kg^{-1}. Values for OG are similar to those reported (127 to 165 g kg^{-1}) by Marten and Hovin (1980); however, they reported lower values for TF (129 to 155 k kg^{-1}). Marley et al. (2014) reported CP values for PRG that were slightly lower, with a mean of 157 g kg^{-1}. Smit et al. (2006) reported values of 155 to 203 g kg^{-1} for six PRG cultivars, and values reported by Ribeiro Filho et al. (2019) ranged from 133 to 191 g kg^{-1}. Interestingly, in previously reported values from Ribeiro Filho et al., PRG-white clover swards had lower CP values than grass only swards, with an average decrease of 17 g kg-1 (2005). Kleen et al. (2010) reported similar CP values for PRG-white clover pastures under beef cow grazing (147 to 161 g kg^{-1}).

Neutral detergent fiber values reported by Allen et al. (2013) ranged from 396 to 547 g kg^{-1}. In both years of the Allen et al. study (2013), similar patterns and trends between OG, TF, MF, and PRG were observed. Smit et al. (2006) reported mean NDF values of 476 to 490 g kg^{-1} for six PRG cultivars. Marley et al. (2014) evaluated PRG under beef steer grazing and reported NDF values of 531 g kg^{-1} which are similar to results from Ribeiro Filho et al. (2005, 2019) who examined PRG and PRG-white clover under dairy cow grazing (507 to 575 g kg^{-1}). Kleen et al. (2010) found similar NDF values (508 g kg^{-1}) under beef cow grazing in Germany. Solomon et al. (2014) reported the overall greatest NDF values, averaging 590 g kg^{-1} for diploid cultivars and 554 g kg^{-1} for tetraploid cultivars. All of these studies are consistent with findings from Marten et al. (1987) that suggest most CSG contain between 500 and 600 g kg^{-1} NDF. Allen et al. (2013) reported NDF digestibility (NDFD) values ranging from 458 to 901 g kg^{-1} for all CSG species in the study, and from 527 to 901 g kg^{-1} for PRG. Solomon et al. (2014) reported similar NDFD values for PRG, averaging 777 g kg^{-1} for diploid cultivars and 807 g kg^{-1} for tetraploid cultivars.

Both DeBoer et al. (2017) and Jaqueth et al. (2019) reported inverted and low calcium (Ca) to phosphorous (P) ratios in annual warm-season grasses, and cool-season turfgrasses, respectively. While intake of both Ca and P is important, the ratio between the two is also important. For most classes of horses, the NRC (2007) recommends a ratio between 1:1 and 6:1. If the ratio is below 1:1, supplementation of Ca is necessary to increase mineral availability for bone metabolism (Caple et al., 1982; NRC, 2007).

Nonstructural carbohydrates are of great importance within the field of horse nutrition. Although NSC are rarely a concern for other livestock species, the horse industry has placed a magnifying glass on NSC as issues with equine obesity and insulin resistance increase. There has not been recommendations or limits established for metabolically normal horses; however, for horses with metabolic issues such as insulin resistance, it is recommended that the total ration contains less than 12% or 120 g kg^{-1} NSC (Frank, 2009). Research on warm-season annual grasses (DeBoer et al., 2017) and legumes (Catalano et al., 2019) indicated that these forages are a potential option for metabolically challenged horses that are otherwise unable to participate in grazing. However, CSG tend to have a slightly larger NSC range. Some data indicates that CSG species are appropriate for metabolically challenged horses, whereas some are above the threshold of 120 g kg^{-1}. For example, Allen et al. (2013) reported NSC values (83 to 169 g kg^{-1}). Perennial ryegrass was within a group of grasses (KBG, MF, and PRG) with greater NSC (104 to 169 g kg-1) compared to meadow bromegrass [*Bromus biebersteinii* Roem. & Schult], OG, and reed canarygrass [*Phalaris arundinacea* L.](63 to 95 g kg^{-1}). Mayland et al. (2000) reported a range of 108 to 138 g kg^{-1} of TF cultivars under cattle grazing. Siciliano et al. (2017) examined the effect of sward height of TF pasture on NSC concentrations. Short swards (15 cm) had lower NSC concentrations (126 g kg^{-1}) compared to tall swards (30 to 40 cm, 163 g kg^{-1}). This difference was attributed to the active growing taking place in the short sward pastures. These values are similar to the TF values reported by Mayland et al. (2000) and Allen et al. (2013). Although maintaining a field at a lower height would likely require more labor, this study demonstrates a viable way to reduce NSC concentrations in existing CSG pastures for horses that may benefit from a reduced NSC diet.

Challenges of Pasture Management in the Upper Midwest

The upper Midwest has cold winters. This is helpful for weeding out the weak, within both plant species and humans. While this is beneficial for population and traffic control, this environment is not conducive to pasture longevity. Although CSG have been demonstrated to establish well, have high yields, animal preference, and nutritive value, they often decrease in productivity after a cold winter. Winterkill and winter injury are often likely the largest factors to the decreased persistence observed in pastures in locations with extreme winters. Currently marketed PRG forage varieties are best adapted to regions with milder winters and lack the winter-hardiness to persist in the Upper Midwest. However, because of its high yield potential and high forage nutritive value, PRG varieties developed by private companies are being marketed in Minnesota. While these varieties do provide short-term advantages compared to other cool-season grasses, they frequently winterkill and productive stands typically last until the year following seeding (Allen et al., 2012; Martinson et al., 2016). Evaluating new cultivars for increased winter hardiness is an important component to providing research-based information to farmers and horse owners in the upper Midwest.

Equine Bodyweight and Body Condition

In general, horses are a much more diverse group compared to other livestock species. The vast number of breeds and disciplines results in a varied range of energy output for horses; therefore, there is not a single ratio of feedstuff that applies to all horses. Horses can be kept at maintenance, where they are seldom (or never) asked to exert energy beyond their free will. These horses have digestible energy (DE) demands ranging from 1.52 to 2.00 Mcal/kg BW (NRC, 2007). Horses can also be Olympic level athletes, competing in strenuous events including three-day eventing and racing. These horses competing at the upper echelons of their discipline are often referred to as elite or performance horses (Pratt-Phillips, 2016). Within this range of workload, there are also horses described as hard keepers or easy keepers. Both types can present management challenges. Hard keepers are horses that are prone to weight loss, regardless of quantity and quality of feedstuffs in the ration (Becvarova et al., 2009). Other classifications of horses with high-energy demands include growing horses, lactating broodmares and breeding stallions. Digestible energy needs for these horses range from 2.18 Mcal/kg BW for a 500 kg breeding stallion, between 2.66 and 3.45 Mcal/kg BW for a 500 kg horse in heavy to intense work, and up to 4.15 Mcal/kg BW for a 3 month old foal, which is the age most foals will start consuming forage and concentrates (NRC, 2007). In contrast, easy keepers are horses that easily gain bodyweight and are thought to be metabolically efficient (Becvarova et al., 2009). It is the easy keeper that is becoming more and more of a management challenge; therefore, this review will focus primarily on the overweight horse (BCS ≥ 7) versus the underweight horse.

Horses prepare for the winter by increasing their forage intake because of an increase in appetite, and by growing a thick coat of hair (Ssewannyana et al., 1990; Donaldson et al., 2004). These changes allow the horse to store energy, which carries them through the winter months when food is scarcer and conditions are harsh, (Johnson et al., 2009). In the wild, most adipose stores have been depleted when spring arrives. This cycle has led to the inheritance of what is known as the “thrifty gene” (Prentice, 2005) and it is thought that thriftiness is related to insulin resistance (IR; Johnson et al., 2009). In a study investigating a specific line of swine (*Sus scrofa domesticus*) that had evolved to have a thrifty genotype due to cycles of feasting and famine found that when these animal were allowed to consume excess food in captivity, they developed excessive adiposity, IR, hypertriglyceridemia, and hypercholesterolemia in comparison to domestic swine (Martin et al., 1973; Buhlinger et al., 1978; Dyson et al., 2005). Horses with the thrifty gene are able to withstand harsher environmental conditions more easily than horses lacking the gene. However, today's horses rarely have to endure the same conditions their wild ancestors did. Although horses still live in cold climates, they are provided shelter, consistent and high quality feed stuffs, and are often blanketed to aid in body temperature maintenance. Under these modern conditions, the thriftiness gene can manifest itself as weight gain. Certain breeds including Morgans, and pony breeds appear to be "thriftier" and are more prone to IR and obesity (Treiber et al., 2006).

Health Parameters Associated with Obesity. There is a growing trend towards obesity of horses in the United States and Europe. Most instances of obesity are the result of an imbalance between energy intake and expenditure (Geor and Harris, 2009). Johnson et al. (2009) observed husbandry practices including feeding of high-energy rations to physically inactive horses. These horses were receiving more calories than they were exerting and as a result, weight gain and fat deposition occurs. Excess BW can lead to health issues including IR and laminitis, both of which can lead to Equine Metabolic Syndrome (EMS; Treiber et al., 2006; Geor and Harris, 2009), poor thermoregulation (Webb et al., 1990; Cymbaluk and Christison, 1990) and decreased athletic ability (Thorton et al., 1987). Pearson et al. (2018) evaluated relationships between body condition score (BCS), body fat, activity level, and inflammatory biomarkers. Overweight and obese horses had body fat percentages ranging from 16.5 to 19.1%, compared to thin and moderate horses, which had body fat percentages of 12.9 to 15.6%. Obese horses had greater plasma prostaglandin E_2, which is a marker of inflammation. This increased inflammation may be a component to the decreased athletic ability, and both the inflammation and the decreased athletic ability are likely due to the strain on the body from carrying "excessive bodyweight". Recently, an interesting study came out examining the effect of increased adiposity on insulin sensitivity (Bamford et al., 2016). Obesity was induced by either a carbohydrate (CHO) rich diet or a fat rich diet. Both groups significantly increased BCS and the CHO group had lower insulin sensitivity compared to the healthy control group; however, the group fed a high fat ration had insulin sensitivity similar to that of the control group (Bamford et al., 2016). This information is useful when considering how to manage metabolically challenged horses.

Some breeds are known to be more predisposed to BW gain, including ponies, Morgans (Norton et al., 2018), and Andalusians (Martin-Gimenez et al., 2016). It has been documented that pony breeds are at an increased risk for obesity (Giles et al., 2014) and insulin resistance (Rijnen and van der Kolk, 2003), and have a higher average BCS compared to light breeds of horses (Pratt-Phillips et al., 2010). In the same study (Pratt-Phillips et al., 2010), Morgan horses also had a higher average BCS compared to Standardbreds and Thoroughbreds. Norton et al. (2018) determined several traits associated with EMS were moderate to highly heritable in both ponies and Morgan horses. Bamford et al. (2014) evaluated breed differences in insulin sensitivity, and found that both Andalusians and ponies had lower insulin sensitivity compared to Standardbreds. These findings emphasize that these breed-types require additional management and care to prevent excessive BW gain and related disorders. By emphasizing the management and tracking of BW, horse owners can minimize potential health risks.

Methods to Assess Body Condition. Detecting changes in adiposity is key to owner management of horse obesity. The most common method to track bodyweight change is to look at body condition, which refers to the amount of subcutaneous fat deposition. Similarly to humans, horses tend to deposit adipose tissue in specific areas. The six areas of interest on the horse include the crest of the neck, the withers, along the topline, behind the shoulder, over the ribs, and the tail head area (Henneke et al., 1983). The BCS system was developed as a way to assess adiposity and places horses on a scale that ranges from one (emaciated) to nine (obese). This is a subjective score and is best used by the owner or a trained professional to track changes over time. The shortcoming with the BCS systems is the loss of sensitivity when BCS approaches the highest scores (≥7 out of 9; Dugdale et al., 2011, 2012). This is likely due to the accumulation of visceral vs. subcutaneous fat. This is supported by data from weight loss studies with demonstrated BW loss without a change in BCS (Dugdale et al., 2012; Glunk et al., 2015). Glunk et al. (2015) proposed that BW loss was the potential result of either visceral fat or reduced gut fill. Although BCS may lose some sensitivity within obese horses, it is still an important tool for both scientists and educators, as well as the horse owner. This system is easily learned and is perhaps the most accessible option for owners to use when evaluating their horse's condition. Where the system is most applicable is the owner using it consistently over time. Even if the scorer is not 100% accurate, if they assess their horse frequently, they will be able to detect subtle changes before fat stores become a health issue. Another easily accessible tool for assessing body condition is the cresty neck score, developed by Carter et al. (2009). The cresty neck score ranges from 0 (no crest) to 5 (crest permanently drooping to one side). Increased fat deposits along the neck of horses and ponies have been shown to be associated with IR and EMS (Carter et al., 2009). Both of these systems look specifically at adipose tissue and it is important to note that accuracy largely depends on knowledge of the scoring system, and repeated assessment of the horse by the same, trained person. In terms of usability, the cresty neck score system is less nuanced than the Henneke BCS system, and is likely easily implemented by owners. Use of this system may aid in earlier diagnosis of IR, in conjunction with other routine veterinary care.

Methods to Determine Body Weight. Using a scale to determine exact BW is ideal; however, most horse owners do not have access to a livestock scale. The two most commonly used options for BW estimation are tapes and estimation equations. Weight tapes are readily available; however, their accuracy has been questioned (Ellis and Hollands, 2002; Wagner and Tyler, 2011; Martinson et al., 2014; Catalano et al., 2016; Jensen et al., 2016). This is in contrast to Pearson et al. (2018) who found no difference between scale weight and weight estimated by a tape in the first 14 horses measured in the study. The majority of horses in that study were stock-type horses. Many of the original BW and condition estimations were based on a 500 kg Quarter horse (i.e. the “average” horse). While these formulas may work well if the horse is moderate in all aspects, they are less applicable for horses that are more extreme, both in physical size and body condition. For many larger breeds, such as warmbloods and draft horses, commonly available weight tapes are not long enough to even estimate BW for those horses. Jensen et al. (2016) evaluated body condition and BW of Icelandic horses and found poor agreement of estimation of BW between two different tapes. If weight tapes are used, care should be taken to select an appropriate tape for the size or breed of the horse.

Bodyweight estimation equations exist for adult light horse breeds (Milner and Hewitt, 1969; Hall, 1971; Carroll and Huntington, 1988), ponies (Owen et al., 2008) and miniature horses (Bruce et al., 2010) utilizing heart girth circumference and body length measurements. Hoffmann et al. (2013) adapted the Carroll and Huntington 1988 equation to more accurately estimate Icelandic horse BW. Growing Thoroughbred horse BW can be estimated by the equation created by Staniar et al. (2004). Recently, new equations were developed that improved existing BW estimation equations by adding breed type, neck circumference and height (Martinson et al., 2014; Catalano et al., 2016). An equation for growing Warmblood horses was also recently developed, and utilizes body length, sternum height, heart girth, and front pelvic width (Coudková et al., 2016). All three of these recent equations reported improved accuracy with additional measurements beyond heart girth and body length, as they appear in the original Hall equation. Age is a poor predictor of BW, due to the rapid growth and BW gain observed in the first year (Coudková et al., 2016). Therefore, developing estimation equations specifically for growing stock within all breeds or types is needed.

Martinson et al. (2014) and Catalano et al (2016) also developed ideal BW estimation equations for draft horses, ponies, Arabians, stock horses, and Warmblood breeds to better equip horse owners and professionals with tools to manage horse BW. This data is supported by a study examining markers for predicting body condition of broodmares. Abo El-Maaty et al. (2017) evaluated Egyptian Arabian mares and found positive correlations between rump fat and BCS and BW, and a negative correlation between the girth to height ratio and thyroid hormones. The ideal BW estimations give owners a tool to use in addition to BCS in determining if their horse is overweight. This information is critical when we consider the data that suggests most owners cannot recognize what ideal looks like. It can be challenging to be subjective when assessing one's own horse, as there tends to be an emotional attachment involved. By having an objective assessment via measurements, the ideal BW prediction gives owners a concrete goal to aim towards.

Horse Owners Management of BW. Despite the fact that the detrimental effects of excess BW have been extensively studied, most owners are maintaining their horses at a higher BW and condition than what would be considered healthy by a veterinarian or nutritionist. It has been well documented that there is a growing population of overweight and obese horses, both in the United States and abroad (Donaldson et al., 2004; Carter et al., 2009; Brooks et al., 2010; Harker et al., 2011; Thatcher et al., 2012; Martinson et al., 2014). Catalano et al. (2016) reported that 42% of draft horses exhibited at county fair were overweight or obese, and Jensen et al. (2016) reported that 24% of mature Icelandic horses in Denmark were overweight or obese. Robin et al. (2015) determined draft and cob-type horses, as well as ponies, were more likely to be overweight compared to Thoroughbreds. Most recently, Jaqueth et al. (2018) determined 40% of the Maryland horse and pony population was over-conditioned. Additionally, there is an increased cost of $434 dollars to manage an over-conditioned horse (Jaqueth et al., 2018). Labor was the primary contributor to the increased cost, along with purchasing and maintaining BW management tools such as muzzles or fencing for dry lots, non-routine veterinary visits, and specialized shoeing. There is not one specific reason why this increase in equine obesity is occurring, although predominant factors include lack of owner awareness and willingness to implement and follow through with BW loss programs (Morrison et al., 2017). Jaqueth et al (2018) agreed, noting that some of these costs could be reduced by educating owners how to best control body condition before it manifests as metabolic disorders. Hitchens et al. (2016) reported that people caring for fewer horses tended to have more horses that were overweight, compared to those caring for a larger herd.

Adding to this problem is that owner perception of horse BW and body condition can be inaccurate. It has been determined that horse owners lacked the ability to detect changes in adiposity over time (Mottet et al., 2009).Wyse et al. (2008) reported an obesity rate of 45% in a group of horses used for recreational riding. Within a subset of horses considered obese by researchers, 37% of owners reported the horse was not overweight. Additionally, of the subset of horses not considered overweight or obese by researchers, 28% of owners considered the horse overweight. Results from Jenson et al. (2016) are in agreement with these findings. There was a low agreement between horse owner and experienced person when assessing BCS, with owners more likely to underestimate BCS of their horse. Morrison et al. (2017) assessed owner ability to determine overweight or obese horses from photographs, with only 11% of respondents correctly identifying an overweight or obese horse. Additionally, there was no difference between professionals (managers and/or trainers) and non-professionals (owners) in their ability to identify an overweight horse (Morrison et al., 2017). Similar results were observed when draft and warmblood horse owners were asked to guess the BW of their horse. On average, owners were 52 kg off the actual BW, with a roughly equal amount of over- and under-estimation (Catalano et al., 2016). The inability of horse owners and professionals to accurately assess BW is likely contributing to the increase in obesity and warrants development of educational materials and tools that can assist with horse BW management. In addition to being aware of excess BW in a horse, implementing a BW loss program can be challenging. Although BW loss is very achievable in a research setting (Glunk et al., 2015), lack of owner compliance is often the biggest setback in an applied setting (Gill et al., 2016). Gill et al. (2016) evaluated the efficacy of 10 to 20% calorie restriction in overweight horses on privately managed farms. The study was conducted over 6 to 7 months and included a wide range of breeds. All but one horse (3 years) was between 7 and 17 years. Overall, horses had a 6% reduction in BW and a 1.4 unit decrease in BCS, with the greatest changes observed in horses whose owners had greatest compliance. One key protocol point of this study was that the researchers worked with individual owners on a case by case basis to implement realistic changes. Many owners board their horses, which can limit their ability to adjust nutritional management strategies. This study created realistic protocols, which likely led to the high compliance rate. These findings are important when working with individuals to create BW loss plans for their horses.

Beyond horse owner and manager awareness of excessive BW is an apparent preference for over-conditioned animals. Society appears to prefer an over-conditioned animal to one that is under-conditioned (Owers and Chubbock, 2013). Previous research (Catalano et al., 2016) discussed a possible show ring preference in which exhibitors were rewarded for having horses with full, smooth and rounded hindquarters, which are often seen in overweight animals. In one study, an overweight pony with a 6.5 BCS was rated as “about right” for showing (Morrison et al., 2017). This preference may be due to a lack of awareness that excessive BW contributes to multiple health risks as previously discussed. Within many livestock species, animals used for reproductive purposes are commonly kept at a greater body condition. Within mares, reproductive success is positively associated with a heavier body condition (Henneke et al., 1984; Fradinho et al., 2014; Morley and Murray, 2014). This common husbandry practice likely has influenced how all classes of horses are kept.

Water for Horses

One often overlooked area of horse husbandry and farm management is water, and specifically, water quality. Water is one of the six key nutrients needed by horses and humans alike. Water is essential for body fluid balance, digestive function, and gastrointestinal health (NRC, 2007). Daily requirements range from 21 to 29 L per day for an idle 500-kg horse on a hay-based diet, although this varies based on a number of conditions including workload and ambient air temperature (NRC, 2007). Water intake occurs directly by drinking fluid water and by consuming feed that contains a high percentage of moisture, and indirectly though metabolism of macronutrients (NRC, 2007). It is generally understood that pasture can provide horses with some water; however, when horses are on a primarily hay-based diet, the majority of their water requirements will be fulfilled through drinking water. Ensuring good water quality is paramount to maintaining overall health in the horse (Hinton, 1978; Jones, 2004; Lardy et al., 2008).

A lack of water, or dehydration, can be very dangerous for a horse. Indicators of dehydration include failure of the skin to return to a flat plane when pinched and held briefly, with hydrated skin returning flat in one second or less. Other indicators include jugular vein refill time, urine volume and color, and gum color. Gums and the inside of the eyelid should be a relatively pale pink (Thomas, 1999). If there is low capillary refill or the membranes are dark red, the horse is likely dehydrated. Brobst and Bayly (1982) examined the effects of dehydration on changes in urine specific gravity and urine osmolality. After 72 hours of water deprivation, horses had lost 13.5% of previous BW. Serum and urine urea nitrogen concentrations increased by 68 and 130%, respectively, and plasma sodium and chloride concentrations increased by 10 and 14%, respectively. Urine sodium and potassium concentrations increased initially but were near normal after 72 hours of deprivation. Azotemia developed in all 6 horses. Although most horse owners are well aware of the need for water, the speed in which dehydration can occur should be emphasized, particularly in the winter when water sources can freeze over quickly, and in the summer in extreme heat.

Water intake and quality preference. Existing equine water-based research has focused primarily on water intake rates, drinking habits in varying conditions, and horse preference. Hinton (1978) discussed water and horses in detail. In terms of requirements, he concluded that water intake is closely related to dry matter intake, and that daily requirements vary based on type of horse, workload, and environmental conditions. Previous to the date of publication (1978), the most commonly used systems for providing water to horses included buckets, bowls in the manger, automatic bowls, troughs, or ponds and flowing water. Large wooden buckets were considered superior to galvanized [steel] or plastic buckets which may be knocked over, dented, or broken. Hinton stressed the important of keeping water containers clean. Today, plastic is the most commonly used material for small buckets, and both plastic and metal are used for larger tanks. The War Office (1904) recommended avoidance of public water troughs, which is still recommended today. If troughs were infected with nasal discharges, they should be emptied, washed with a hot solution of washing soda, and left for 24 hours before rinsing and refilling (Miller, 1965). Although disinfecting technology has advanced, the process is still somewhat similar. Other information regarding timing and amount of water offered is discussed in Hinton's review; however, he concludes that the recommendations that are discussed are often confusing, illegal, or incorrect. Most of the problems described (i.e. colic) probably stemmed from mismanagement and over restricted water rations. Hinton concludes by stating that the most logical advice is to offer horses water in excess of their requirements at rest, and to allow them to drink frequently when working. If water must be rationed, it is advised to water the horse before feeding, and in instances of excessive thirst, the water should be rationed only initially (Hinton, 1978).

Some research has been done on the mineral content, source, and temperature of water preferred by horses. There is mixed data on whether soft or hard water is better for horses, and Hinton (1978) concluded either are acceptable, as long as the water is clean and wholesome. Hinton also concluded that water temperature does not have a great impact on horse intake. Kristula and McDonnel; however, reported that ponies drank 40% more warm water (50°C) than ambient near-freezing water during cold winter weather (-7 to 5°C; 1994). In a follow up study, the same researchers determined that there was no difference in intake of either ambient warm (17 to 31°C) versus ice-chilled (0 to 1°C) water by ponies during hot summer weather (15 to 29°C; McDonnell and Kristula, 1996). Daily water consumption was similar in both studies, despite differences in environmental condition. One limitation to these studies was the fact that water treatments were not offered simultaneously. As such, true preferences were unable to be determined between water temperature ranges. The results of McDonnell and Kristula's work are most impactful when considering water intake in the winter. Providing warm water may reduce the risk of dehydration complications such as impaction colic. Rucker and Hiney (2013) examined seasonal water intake between horse fed dry grain and a soaked mash. They found that horses consuming the mash drank similar or greater amounts of water compared to horses consuming dry grain. This indicates that grain mashes, or other similarly soaked rations, may help increase total water consumption. Pagan et al. (2017) reported an increased water intake (4 to 5%) for every 1°C increase in temperature for Thoroughbred racehorses in training. Gordon and Jerina (2013) also reported increased water intake with increasing temperature, and also reported greater intake when supplemented with a "no taste" salt supplement top dressed over grain rations, compared to voluntary consumption of traditional salt blocks. Additional research has determined that horses have an aversion to acidic (sour) water (Merkies and Carson, 2011).

Horse water source preference. Research has also been conducted on the source of water and the resulting behavior and/or preference of the animal. Van de Weerd et al. (2012) investigated the use of buckets, automatic waterers, and troughs by unhandled semi-feral ponies. Ponies preferred buckets or troughs over the automatic waterers, which researchers theorized was due to the noise during filling. Interestingly, when unfamiliar ponies were added to the established group of three, these preferences did not exist. Researchers theorized that ponies were potentially physically obstructed from drinking from the originally preferred water sources, and chose to face their fears of the automatic waterers over interacting with unknown ponies. Krawczel et al. (2006) evaluated four different models of automatic waterers among naïve horses. Two models using push valves, which requires the horse to exert force on a lever to dispense water, and two models using float valves, which uses an automatic float valve to maintain water at a constant level, were used. There was nearly no consumption from either push valve model, and there was a trend for greater water consumption from the float valve model that made the least amount of noise when refilling, and that had the larger bowl (relative to the other model). This trend was confirmed by individual horse trials in the same study. This preference for a larger surface area has also been observed by horses drinking from buckets or pressure valve bowls (Nyman and Dahlborn, 2001) and by dairy cows drinking from troughs with varying depths and heights (Teixeira et al., 2006). Akam et al. (2017) evaluated the effect of bucket placement on water intake. Buckets were placed either near the stall door or near the feeder. Horses drank more from buckets when they were placed further away from their feed source. This is useful data, and is particularly applicable for those with messy eaters. By placing the bucket away from the eating area, some labor may be mitigated. The same research group also evaluated the effect of bucket color on water intake in horses (Davis et al., 2017). Yellow ("light") and black ("dark") buckets were hung side by side in the front of the stall, rotating every day for six days. No difference was found between either bucket color.

Water Quality

It was not until the mid-20th century that water quality for humans and livestock became of great importance. The 1972 Environmental Studies Board included the memorable quote that it [is], "not necessary to weigh with tenderness and care the testimony of experts… an ordinary mortal knows whether water is fit to drink". This statement comes from a decision with a state supreme court in 1904 (Environmental Studies Board, 1972). Water quality standards, which determine the suitability for consumption, vary widely (Markwick, 2007; NRC, 2007).

Total Dissolved Solids. The primary parameter for assessing water quality for livestock use is the total dissolved solids (TDS) value. Other parameters include odor, color, temperature, and turbidity. Total dissolved solids is a measure of the aggregate composition of the ions in the water (NRC, 2007). Total dissolved solids values under 1,000 mg/L are considered safe and should pose no health risks. Values between 1,000 and 2,999 mg/L are generally safe, but may cause temporary diarrhea in unaccustomed horses. Values above 3,000 mg/L may negatively affect water intake, while water with values above 5,000 mg/L should be avoided for pregnant or lactating animals (NRC, 2007). Water with TDS values above 7,000 mg/L should not be offered (Environmental Studies Board, 1972). However, TDS is generally low (< 350 mg/L) in North American water sources (NRC, 2007). Well water in a study conducted in New Mexico had TDS values of 276 ppm (Mars et al., 1992).

Turbidity. Turbidity is the measurement of scattered light that results from the interaction of light with suspended and undissolved material in a water sample (Lambrou et al., 2009), or more simply put, the relative clarity of a liquid. It is commonly reported in Nephelometric Turbidity Units, or NTU. Human drinking water should have a turbidity of ≤ 5 NTU (Davis and Lambert, 2002), although the World Health Organization (2004) recommends water be below 0.1 NTU for effective chlorine disinfection. Chlorine can still be effective up to 20 NTU (Davis and Lambert, 2002); however, the greater the NTU, the greater the risk of chlorine by-products forming by interacting with the organic matter (Laurent, 2005). In general, water will need treatment to reduce turbidity before disinfection can take place (Wisner and Adams, 2002). Solids in drinking water can support the growth of harmful microorganisms, and additionally, are thought to provide "shelter" for these microbes by reducing their exposure to disinfectants (Lambrou et al., 2009).

Although little data exists on the connection between water turbidity and livestock health, there is a substantial amount of research on the impacts of turbidity on fish. Rainbow trout (*Oncorhynchus mykiss)* were less reactive with increasing turbidity (Barrett et al., 1992), with worsening reaction distances at 15 and 30 NTU compared to ambient (4 to 6 NTU). Bluegills (*Lepomis macrochirus*) had decreased feeding rates with increasing turbidity (60 to 190 NTU; (Gardner, 1981). Along with Coho salmon (*Oncorhynchus kisutch*), Rainbow trout BW increased faster in clear water than in turbid water (Sigler et al., 1984). Additionally, fish in turbid channels (fish rearing system) left the channel to move into migration traps more frequently than fish in clean water. Sigler et al. (1984) hypothesized this difference was due to the stress induced by the turbidity, not from a lack of ability to find food. In this study, the researchers initially used turbidity levels of 100-300 NTU; however fish either all left the channels or died. The turbidity was subsequently adjusted to 25 to 50 NTU, and at 50 NTU, visibility was limited to 2 to 5 cm (Sigler et al., 1984). In contrast to these studies, there are also reports of increased turbidity providing protection to salmon (*Oncorhynchus* spp.) in naturally occurring bodies of water (Lloyd, 1987; Gregory and Levings, 1998). A statewide survey of Illinois streams and rivers reported a median turbidity value of 36 NTU during peak/high water flow and a mean turbidity value of 18 NTU during base flow (Royer et al., 2008)

Within livestock, Fraser et al. (1998) found positive correlations between turbidity and both livestock fecal coliform discharge and concentration levels, confirming that livestock waste has a negative impact on water quality. A clear correlation has also been observed in human health. Schwartz et al. (1997, 2000) found a correlation between increasing water turbidity and hospitalizations of the elderly and young children in Philadelphia, even though NTU levels were very low (<0.3). Contaminants in drinking water can be from multiple routes of exposure; including ingestion, dermal, and even inhalation (Lambrou et al., 2009).

Algae and Chlorophyll *a*. Algae are a growing problem in both naturally occurring and man-made bodies of water, including stock tanks commonly used to provide water to horses and other livestock. Of the contaminants that affect water quality, algae are some of the more visible ones and can be a signal to clean the water source. Algae are photosynthesizing organisms that make their own food by using solar energy, and under the right temperature and water conditions, can grow very rapidly and form extremely high-density populations or blooms (Sheath and Wehr, 2015). Algae blooms can turn the water green and affect odor, which may negatively affect intake rates (NRC, 2007). Specifically, blue-green algae have been documented to decrease water intake (Falconer, 1999). Blooms can also potentially be toxic and pose serious health risks to people, pets, and livestock causing gastrointestinal diseases and death (Beasley et al., 1989; Stewart et al., 2008; Sheath and Wehr, 2015). Falconer (1999) highlighted that the populations at risk are those livestock that have no other water choice, and humans of a lower economic class that may not have access to an alternate water source, or may not be able to afford bottled water.

Algae occur in multiple forms; sestonic cells, epilithic biofilms, and filamentous mats (Royer et al., 2008). Sestonic (water column, versus benthic [bottom]; (Paul et al., 2017)) chlorophyll *a* is an indirect measure of algae abundance (Migliaccio et al., 2007). Chlorophyll *a* responds to both macronutrient loading and availability; therefore, it is thought to be a more sensitive and relevant indicator of water quality than nutrient concentrations (Boyer et al., 2009). The median sestonic chlorophyll *a* value for Illinois streams and rivers in 2004 was 5 mg m^3 (Royer et al., 2008). When canopy cover was ≥ 25%, chlorophyll *a* levels decreased (Royer et al., 2008). This data is applicable for horse owners deciding where to place water tanks, and indicates shade will benefit water quality by limiting algae production. Interestingly, there was no correlation between phosphorus levels and chlorophyll *a* levels; the authors hypothesized that this was due to washout from water flow, compared to ponds or lakes which are more static. Sestonic chlorophyll *a* values for the War Eagle Creek watershed in Arkansas were similar to the previous study, ranging from 0.5 to 6 mg m^3 (Migliaccio et al., 2007). In a man-made reservoir in Taiwan, over a five year timespan, chlorophyll *a* values ranged from 1.71 to 19.83 mg m^3 (Lu et al., 1999), while values from the Chesapeake Bay of varying salinity all had a mean value of 15 mg m^3 (Dennison et al., 1993). Lakes across Minnesota ranged widely in chlorophyll *a* concentrations; ranging from 2.1 to 279 mg m^3 (Brezonik et al., 2005). Lakes with greater chlorophyll *a* had lower clarity; turbidity ranged from ≤ 5 to 155 NTU, (Brezonik et al., 2005). Turbidity, chlorophyll *a*, and total suspended solids were all strongly, positively correlated with each other ($r > 0.95$) whereas all were negatively correlated with Secchi disk transparency ($r > -0.95$; (Brezonik et al., 2005). In contrast to these results, Lloyd el al. (1987) found that when controlling for phosphorous, the nutrient that is typically limiting for algae production, chlorophyll *a* concentrations decreased with increasing turbidity (Lloyd et al., 1987). This is due to the reduced light availability due to particles blocking the UV rays. Although this intuitively makes sense, in general, we can assume that the clarity of most lakes is controlled by algae and algal-related suspended matter.

Other Issues with Water. Strains of *E. coli* can survive in water tanks for up to four months, and can replicate to infectious doses during summer months (Hancock, 1998). Hancock (1998) emphasizes that traditional means of controlling infectious agents such as eradication or test and removal of carrier animals are not feasible for *E. coli*, and that farm management practices are key to reducing the prevalence of this agent.

At high levels, nitrate (NO_3) can pose a health risk for both humans and livestock. In particular, infants are particularly susceptible to nitrate poisoning, which affect oxygen carrying capacity of the blood. This is known as methemoglobin, or "blue baby syndrome" (Sallenave, 2017). Ruminants, horses, and other young monogastrics are also affected by nitrate in a similar manner due to the presence of nitrogen-converting bacteria in their gastrointestinal systems. Nitrate poisoning can be a result of feed, water, or both. The most common issues of nitrate poisoning for horses via feed would be certain forages harvested under drought or frost conditions (DeBoer et al., 2017). Key symptoms include a bluish or brownish discoloration of non-pigmented skin areas and mucous membranes, sluggish gait, staggering, labored breathing, rapid heartbeat, frequent urination, and ultimately collapse (Sallenave, 2017). Currently, there is not a regulatory water quality standard for nitrate for livestock; however, the authors suggest a maximum level of 100 ppm of nitrate-nitrogen (Sallenave, 2017).

Fish

Husbandry. Goldfish are notoriously hardy and known to survive a wide temperature range (Ford and Beitinger, 2005). Fry et al. (1942) reported lower and upper lethal temperatures of 0 and 41°C, respectively, for goldfish. Ford (2005) reported similar results, with a minimum temperature of 0.3°C and maximum temperature of 43.6°C for goldfish. Yoshitomi et al. (2002) evaluated water quality on physiological functions in goldfish. Goldfish were stocked at a rate of 2.8 L per fish, and fed at 1% BW per day. In the control group, a 50% water change occurred once per week; however in the experimental group, goldfish were reared without changing the water for the duration of the 20 week period. In the experimental group, total ammonia nitrogen, nitrate, and phosphate concentrations increased throughout the period; however nitrite levels remained constant. Although the immune, respiratory, and reproductive systems were negatively affected, there were no goldfish mortalities (Yoshitomi et al., 2002).

Fish Diet. Algae represent an important and valuable food source for many aquatic animals, and contain nearly all essential nutrients, including polyunsaturated fatty acids, vitamins, and minerals (Khatoon et al., 2010). Goldfish are commonly fed diets that contain approximately 50% CP, 11-22% fat, and 20% carbohydrates (Abi-Ayad and Kestemont, 1994), and algae have been documented to contain slightly more protein and minerals (Khatoon et al., 2010). In an ongoing effort to reduce the population of feral goldfish in Australia, Morgan and Beatty (2007) evaluated the stomach contents of 20 goldfish that were between 28 to 386 mm in total length. The contents were comprised mainly of blue-green algae, but also contained other algae, various insect larvae, and some terrestrial insects (Morgan and Beatty, 2007).

Fish Impact on Water. It initially sounded very promising that goldfish were found with algae in their stomachs. However, Morgan and Beatty (2007) also included that the stomach of goldfish may in fact give nutrient enrichment to the algae cells or colonies and likely stimulate significant growth of blue-green algae. Furthermore, the bottom feeding action of goldfish may re-suspend nutrients, making them available to algae. This is very counter-intuitive, particularly to those farm owners who use goldfish to control algae. Goldfish have also been demonstrated to increase turbidity. In their study examining the effect of goldfish on controlling algae in urban drainage systems, Scholz and Kazemi-Yazdi (2005) reported higher NTU values in ponds planted with aquatic vegetation and lower NTU values in unplanted ponds after goldfish were introduced. This is likely due to fish foraging on the vegetation, and thus stirring up sediment. This is in agreement with data from Richardson et al. (1995) that determined turbidity increased from a baseline value of 3.3 NTU to 117 NTU after fish were introduced to a mud bottomed pool. Pools with gravel bottoms did not have a significant increase in turbidity from baseline, and would be more representative of a horse tock tank. The increased turbidity likely benefitted the fish, as suggested previously for wild fish. During this study, a single avian predator entered the enclosure and ate all 48 fish from the clear gravel-based pools, while only managing to eat about 50% of the fish from the turbid pools. Goldfish in this study were mainly herbivorous (75%), although they also consumed mud, insects, and small crustaceans (Richardson et al., 1995).

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CHAPTER TWO: YIELD, PERSISTENCE, FORAGE NUTRITIVE VALUE, AND PREFERENCE OF WINTERHARDY PERENNIAL RYEGRASS UNDER LIVESTOCK GRAZING

Summary: Perennial ryegrass (PRG) is a high quality and yielding forage that, until recently, has lacked the winterhardiness to persist in the upper Midwest. The objective was to evaluate the yield, persistence, forage nutritive value, and animal preference of a new winterhardy PRG cultivar ('Forageur') grown in monoculture and in mixture with white clover under horse and cattle grazing. Research was conducted in St. Paul (SP) and Grand Rapids, MN (GR). Meadow fescue (MF), tall fescue (TF), orchardgrass (OG), and three perennial ryegrass varieties ('Forageur', 'Remington', and 'Spreader IV') were seeded in 2015 (SP) and in 2016 (GR); and were grazed for 2 days by either 6 adult horses (SP) or by 6 cow-calf pairs (GR). Prior to grazing, forage yield and quality samples were collected, and post-grazing, fields were visually assessed for the percentage of forage removal to determine livestock preference. Yields of PRG cultivars were similar but lower yielding (6.9 to 7.6 Mg ha^{-1}) compared to other CSG. Persistence was similar among all CSG in SP; however it was lower for PRG in GR. 'Forageur' was highly preferred by both horses (≥74% removal) and cows (≥70% removal) and was consistently among the highest for crude protein (200 g kg^{-1}), equine digestible energy (2.35 Mcal/kg), and bovine metabolizable energy (2.6 Mcal/kg), moderate for nonstructural carbohydrates (136 g kg^{-1}), and among the lowest for neutral detergent fiber (518 g kg^{-1}). This research confirms that 'Forageur' is viable PRG cultivar to use in upper Midwest livestock pastures.

Introduction

Cool-season perennial grasses (CSG) are the foundation of productive pastures in the upper Midwest (Allen et al., 2012), providing year-round ground cover and offering an affordable form of forage for a diversity of livestock. In particular, beef cattle and horses depend on pasture to provide the forage during the growing season. Producers select forage species based on a number of factors including yield, persistence, animal preference, and nutritional content (Rojas-Downing et al., 2018). Allen et al (2012) found that orchardgrass (*Dactylis glomerata* L.; OG), meadow fescue (*Schedonorus pratensis* Huds.; MF) and tall fescue (*Schedonorus aruninaceus* Schreb.; TF) were the highest yielding and most persistent grasses under horse grazing. Casler et al. (2008) in Wisconsin found that under a frequent harvest regime, TF and OG had greater yields compared to MF.

Perennial ryegrass (*Lolium perenne* L.; PRG) is widely grown in regions of the U.S with milder winters where it is identified for its high seedling vigor, forage yields and high forage quality (Rojas-Downing et al., 2018). PRG was consistently high in nutritive value (Smit et al., 2006; Allen et al., 2013; Marley et al., 2014; Ribeiro Filho et al., 2019). In both monoculture and in mixture with white clover (*Trifolium repens* L.; WC), PRG is a highly preferred forage species (Smit et al., 2006; Allen et al., 2013). Throughout multiple experiments evaluating CSG tolerance under horse grazing, PRG was consistently among the most preferred species (≥ 5.7 on a 9 point scale; Hunt and Hay, 1990; Casler and Kallenbach, 2007; Olson et al., 2016). Currently marketed PRG forage cultivars are best adapted to regions with milder winters and lack the winterhardiness for long term persistence in the Upper Midwest. Brummer and Moore

(2000) evaluated perennial cool-season grasses under continuous grazing by beef cattle in Iowa; grasses were not grazed until the first production year; however, PRG did not survive (9% persistence) the first winter and was eliminated from the study. In a comparison of commonly grown CSG species, PRG was moderately ranked for persistence after two years; however, it had a reduction in yield from the seeding year to the first production year of about 26% (Allen et al., 2012). Elgersma et al. (1998) evaluated PRG-white clover mixtures under mechanical harvest in the Netherlands, and reported both winterkill and winter injury in all plots when winter temperature and precipitation was below average.

In addition to being grazed as monocultures, perennial CSG are often grown in mixture with legumes such as white clover. Mixtures provide many advantages associated with species diversity such as enhanced resource utilization and insurance of some stand productivity should one of the species in mixture fail to persist (Sheaffer et al., 2018). Specifically, WC is often added to pastures as a way to increase forage nutritive value and to optimize the benefits of nitrogen fixation (Collins and Rhodes, 1989). It is low growing, can tolerate frequent defoliation and perenniates through stolons and reseeding (Sheaffer et al., 2018) A positive impact of clover on total forage yield was reported by Elgersma et al. (1998). Yields for PRG-WC systems ranged from 10.8 to 14.6 t ha^{-1} under dairy cow grazing (Schils et al., 1999).

Ryegrass has potential as a high quality forage but marketed cultivars lack sufficient winterhardiness for long-term productivity. The PRG breeding program at the University of Minnesota has identified several potential new cultivars with improved winterhardiness, persistence under frequent mowing, and increased forage yield. These characteristics are desirable in the forage industry where few pasture grass cultivars have been released and even fewer have been evaluated in cold climates or under grazing. Therefore, the objective was to evaluate the forage yield, stand persistence, forage nutritive value, and animal preference of a new winterhardy PRG cultivar (PRG 'Forageur') grown in pure stand and in mixture with white clover under horse and cattle grazing.

Materials and Methods

Research was conducted using horses in St. Paul (SP, 44°59'14"N, 93°10'37"W) and beef cow-calf pairs in Grand Rapids, MN (GR, 47°14'26"N, 93°31'17"W). The SP experiment was seeded in 2015 on a Waukegan silt loam (a fine-silty over sandy or sandy-skeletal, mixed, superactive, mesic Typic Hapludoll). The GR experiment was seeded in 2016 on an Itasca silt loam (a coarse-loamy, mixed, superactive, frigid Haplic Glossudalfs). Soil pH, K, and P were adequate at both locations for legume and cool-season grasses according to Minnesota fertility guidelines (Kaiser et al., 2011). For both experiments, the experimental design was a randomized complete block with four replicates. Treatments were arranged in a split plot design. Whole plots were legume treatment and included CSG grown in monoculture (monoculture) or in mixture with WC 'Kopu II'. Subplots were cool-season grasses and included MF 'HDR', TF 'STF-43', OG 'Intensiv', and three PRG cultivars; 'Forageur' [test cultivar], 'Remington', and 'Spreader IV'. Plots were established using a small-plot drill on 11 Sept. 2015 (SP) and 22 Aug. 2016 (GR). Individual plots were 1.8 m wide by 6.1 m long. All grasses were seeded at a rate of 28.0 kg ha^{-1}, with the exception of OG, which was seeded at a rate of 13.5 kg ha^{-1}. White clover was seeded at a rate of 3.4 kg ha^{-1}. A selective broadleaf herbicide (dicamba [3,6-dichloro-2-methoxybenzoic acid] at 0.19 L ha^{-1}) was applied once in the spring of 2016 to the monoculture grass plots in SP to control weeds. No chemical weed control was necessary in GR. Grass monoculture plots were fertilized with 56 kg N ha^{-1} (urea) in early May of each year.

For both experiments, the first grazing of each year was initiated when the majority of grasses were in the early elongation stage (Moore et al., 1991) and subsequently followed a 28-day grazing interval after the first grazing period, which allowed for regrowth to reach about 20 cm. Forage yields were determined by mechanically harvesting a 0.9 by 5 m area to 8 cm using a flail harvester (Carter Manufacturing Company, Inc., Brookston, IN). The harvested area accounted for half of each seeded plot and the remainder was used for grazing and determination of preference. At each harvest, the areas used for mechanical harvest and grazing were rotated as described by Allen et al. (2012). The SP experiment was harvested for yield on 31 May, 27 June, 25 July, 29 Aug., and 10 Oct. 2016 and on 5 June, 3 July, 31 July, and 11 Sept. 2017. The GR experiment was harvested for yield on 12 June, 17 July, 14 Aug., and 19 Sept. 2017 and on 4 June, 2 July, 30 July, 27 Aug. and 26 Sept. 2018.

Species stand persistence was measured using a frequency grid in two locations per plot (Vogel and Masters, 2001) after the last harvest of the seeding year, prior to the first harvest of the first production year, after the last harvest of the first production year, and finally in the spring following the final harvest the previous fall. Plant height was taken at five locations within each plot and averaged. Grass maturity was determined using the scale established by Moore et al (1991). Clover maturity was not determined because it was consistently in a vegetative stage throughout all grazing events.

Samples for determination of botanical composition, dry matter content, and nutritive value were collected from three representative 0.09 m^2 areas to 5 cm from each plot prior to grazing. Botanical composition of mixture plots was determined by separating these samples into legume and grass components. Each component was weighed and then recombined for drying and forage nutritive analysis. Samples were kept below ambient temperature during sorting to minimize any changes in nutritive value. All samples were dried at 60°C for 48 hours and weighed to determine dry matter (DM). Samples were then ground through a 6-mm screen in a Wiley mill (Thomas Scientific, Swedesboro, NJ) followed by a 1-mm screen in a Cyclotec (Foss, Eden Prairie, MN). Samples were mixed thoroughly and submitted for forage nutritive analysis using near-infrared spectroscopy at a commercial forage testing laboratory (Dairy One Labs, Ithaca, NY). Crude protein was calculated at the percentage of N multiplied by 6.25 (method 990.03; (AOAC International, 2010)). Neutral detergent fiber was measured using filter bag techniques (Ankom Technology, 2013) and starch and WSC were measured using techniques described by Hall et al. (1999), with NSC estimated as the sum of WSC and starch (Longland and Byrd, 2006). Mineral concentration were determined (Thermo Jarrell Ash IRIS Advantage HX Inductively Coupled Plasma Radial Spectrometer, Thermo Instrument Systems Inc., Waltham, MA) after microwave digestion (Microwave Accelerated Reaction System, CEM, Mathews, NC). Equine DE was calculated using an equation developed by Pagan (1998) and cattle ME was calculated using the NRC equation (NRC, 1988).

Following harvest for yield and forage quality, plots were grazed by six adult horses (526 kg ± 30, 5.5 ± 0.8 body condition score; Henneke et al., 1983) and by four to six beef cow-calf pairs at SP and GR, respectively. Stocking rate was adjusted to allow for biomass removal in two days. Preference ratings were taken after four hours after initiating grazing for both experiments. Livestock were removed when the majority of grasses had been grazed to an average height of 10 cm. Livestock were offered ad libitum access to water while grazing. Preference was measured by visually assessing the percentage of available forage removal on a scale of 0 (no grazing activity) to 100 (100% of available forage grazed; Marten et al., 1987; Allen et al., 2013). In SP, manure was manually removed at the end of each day, and once grazing was completed, stands were mowed to a stubble height of 8 cm to allow for even regrowth. Manure removal and mowing were not done post-grazing in GR due to more consistent cattle grazing and less solid manure piles. When not grazing, livestock were either maintained on a dry lot with grass hay or in an alternate grass pasture, both with ad libitum access to water. Horses were also fed 2.2 kg of ration balancer (Nutrena Empower Balance, Elk River, MN) daily to ensure trace minerals and vitamin needs were met (NRC, 2007). All experimental procedures were conducted according to those approved by the University of Minnesota Institutional Animal Care and Use Committee (beef cow: 1704-34737A; horse: 1603-33539A).

Data were analyzed using the MIXED procedures of SAS (version 9.4; SAS institute, Cary, NC). Response variables included forage yield, maturity, height, grass:clover proportion, forage nutritive values (CP, NDF, Ca, P, NSC [horse only], horse DE, cattle ME, NDFD at 48 hours [beef cow only]), and preference (percent removal). Experiment (location), year, grass treatment, and clover treatment, along with all interactions, were evaluated as fixed effects and replicate was included as a random effect. Maturity was evaluated as a covariate for all models. Individual plots were the experimental unit and statistical significance was set at $P \leq 0.05$. Correlations were run between yield, height, maturity, preference, and all forage nutritive values. Yield is presented as season total (Mg ha^{-1}). All other variables are presented as weighted season means. Means are the least square means of the MIXED procedure (±SE), and mean separations were determined using Tukey's test ($\alpha = 0.05$).

Results and Discussion

Weather

Average air temperatures were typically near historical averages for the duration of the experiment (Figure 1A-B). Noteworthy exceptions include a warmer than average May in 2018 in GR and a cooler than average Oct. in 2018 in GR (Figure 1B). In SP, precipitation was typically above average, particularly in May 2017, July 2016, August 2016 and 2017, and September 2016 (Figure 1C). Grand Rapids also received more precipitation than average in 2016 and 2017 (Figure 1D).

Forage yield

Forage yield was affected by location, year, grass, and the year by grass interaction as fixed effects ($P \leq 0.0402$; Table 1). Inclusion of white clover as a mixture component did not affect yield ($P = 0.198$). Overall, forage yields were greater at GR (10.2 Mg ha^{-1}) compared to SP (6.1 Mg ha^{-1}; $P < 0.0001$) and year 1 yielded more forage (10.4 Mg ha^{-1}) than year 2 (5.9 Mg ha^{-1}; $P < 0.0001$) in both experiments ($P < 0.0001$). All three PRG cultivars were lower yielding (6.9 to 7.6 Mg ha^{-1}) than OG, MF, and TF (8.6 to 9.3 Mg ha^{-1}), with the exception that PRG 'Remington' was similar to TF (table 2) However, the grass by year interaction occurred primarily because the three ryegrasses had similar yields as MF, OG, and TF in year 1 but lower yields than these other grasses in year 2. The experimental cultivar, PRG 'Forageur' was among the middle-ranked cultivars in the seeding year (9.8 Mg ha^{-1}) and among the lowest (4.4 Mg ha^{-1}) in year 2. The three PRG varieties were similar in yield for both year 1 (9.5 to 10.2 Mg ha^{-1}, $P \geq 0.05$) and year 2 (4.2 to 4.4 Mg ha^{-1}, $P \geq 0.05$).

Results of the current study are similar to yield reported by Allen et al. (2012) which ranged from 8.7 to 9.2 Mg ha^{-1} for MF, 9.2 to 12.6 Mg ha^{-1} for OG, 6.6 to 9.0 Mg ha^{-1} for PRG, and 8.4 to 10.1 Mg ha^{-1} for TF when grasses were grown in Minnesota. Yields for OG and TF were similar to results reported by Marten and Hovin (1980) which ranged from 7.2 to 11.0 Mg ha^{-1} (OG) and 5.4 to 7.0 Mg ha^{-1} (TF) in a 4-cut system. Yields for TF and MF were similar to means reported by Casler and van Santen (2001) which ranged between 8.3 and 8.6 Mg ha^{-1} under cow and heifer grazing. Yields of PRG were considerably greater compared to studies completed in both Mississippi (Solomon et al., 2014) and the United Kingdom (Gibb and Baker, 1989; Orr et al., 2005; Marley et al., 2014), which reported yields ranging from 1.3 to 3.9 Mg ha^{-1}. Those studies utilized rotational grazing and/or careful management of post-grazing height; however, the explanation for the relatively low yield is unclear. Kleen et al. (2010) reported more comparable yields between 7.2 and 10.6 Mg ha^{-1} of PRG-white clover mixtures; however, Wilman and Asiegbu (1982) reported lower yields of PRG-white clover mixtures on a 4-week harvest schedule (3.5 to 4.1 Mg ha^{-1}).

Yield Composition

There was a location by year interaction for clover composition of the mixtures ($P \leq 0.0012$). However, there was there was no effect of grass species or cultivar on G:C ($P =$

0.2889). Overall, the proportion of clover in the mixtures was small with an average G:C of 9:1 at St. Paul and 82:1 at GR. In SP, G:C was similar in year 1 (8:1) and year 2 (11:1), but at GR, clover composition was much lower in year 1 (139:1) compared to year 2 (25:1). Wilman and Asiegbu (1982) reported average clover proportions of PRG-WC swards that were considerably higher than those observed in the current study with a mean of 17.1% clover (roughly 5:1). They did; however, report that clover content was very low in the seeding year, and clover increased by lengthening the time between harvests (Wilman and Asiegbu, 1982). This is in contrast to other work within both horse (Catalano et al., 2019) and dairy cattle (Schils et al., 1999) rotational grazing systems which reported that white clover content decreased over time. Catalano et al. (2019) reported a change in white clover to grass ratio from 5.1:1 in the seeding year to 0.8:1 in the first production year, and Schils et al. (1999) reported that the grazing activity of the dairy cows reduced clover content by 12% compared to the cutting control.

Forage Persistence

Persistence was evaluated using the final ratings, taken the spring following the final harvest for each location. Location, clover, location by grass and location by clover interactions all affected persistence ($P \leq 0.0001$). In SP, there were no differences in persistence among grasses (≥ 63%; Fig 2) but at GR, 'Spreader IV' and 'Forageur' had less persistence than MF, OG, and TF (50 to 82%). The location by clover interaction occurred because…

It is well documented that PRG is less persistent than other cool-season grasses (Brummer and Moore, 2000; Allen et al., 2012; Olson et al., 2016); however, we are uncertain why 'Forageur' was numerically lower in persistence compared to the other two PRG cultivars in both SP and GR. During January 2019, the upper Midwest endured a 9-day stretch of unusually cold temperature where overnight lows ranged from -26 to -42°C. Although there was snow cover at this time, this likely contributed to the increased winter kill and injury observed in GR compared to SP.

Forage Height and Maturity

There were many treatment interactions affecting both variables, therefore an in depth discussion is not warranted except as they correlate to forage yield and forage nutritive value parameters. Height was affected by location by year, location by grass, location by clover, year by grass, year by clover, location by year by grass, and location by year by clover interactions ($P \leq 0.005$). Plant height ranged from 29 to 61 cm (Table 2). In year 1, OG tended to be taller than all other grasses. All three PRG varieties were the shortest in year 1, and in year 2, although in year 2, PRG 'Remington' was similar to year 2 TF. As expected, height was positively correlated with yield ($R^2 = 0.58$; $P < 0.0001$). The final model for plant maturity included location, year, grass, and all interactions within those effects ($P < 0.008$). At the time of harvest and grazing in both years, all varieties were in the elongation stage (2.1 to 2.6; Moore et al., 1991; Table 2) with the exception of OG in year 1, which remained in a vegetative stage (1.8). Heights for PRG are similar to previously reported values of 25 to 30 cm under dairy cow grazing (Ribeiro Filho et al., 2019). Grass maturities in the current study were similar to those reported by Allen et al. (2012), which were maintained on a similar grazing schedule.

Forage nutritive values

There were many significant main treatment effects and interactions for nutritive value variables. Averaged for years, location and clover treatments, grasses species/cultivars differed for all variables, while clover affected all variables except P concentration. The most consistent interaction effect was the year by grass interaction and in the following discussion we will focus on this interaction. Year by clover interactions often occurred because of the varying clover content within years and locations. Significant effects within the forage nutritive parameters, a test of all fixed effects for each parameter is included in Table 1. Only significant effects or interactions were included in each model with the exception of CP, Ca, P, and cattle ME. In these four models a main effect was included in the model, even if the corresponding *P* value was > 0.05, if an interaction term of that main effect was significant.

Crude Protein

Overall, PRG 'Spreader IV' had the greatest concentration of CP (214 g kg^{-1}) compared to all other grasses. Tall fescue and PRG varieties 'Forageur' and 'Remington' contained moderate concentrations of CP (195 to 200 g $kg^{-}1$), whereas MF and OG contained the least CP (179 to 180 g kg^{-1}; $P < 0.0001$). Plots with clover contained more CP (202 g kg^{-1}) compared to monoculture grass plots (187 g kg^{-1}; $P < 0.0001$). Comparing across the year by grass interaction ($P < 0.0001$; Table 4), PRG 'Spreader IV' contained more CP in year 2 (224 g kg^{-1}) than in year 1 (205 g kg^{-1}). In year o1ne, PRG 'Forageur' contained 191 g kg^{-1} and was moderately ranked; however, in year 2, it contained among the most CP (209 g kg^{-1}). Crude protein had a positive correlation ($R^2 \geq 0.44$; $P < 0.0001$) with all other forage nutritive values except NDF, in which it had a negative correlation ($R^2 = –0.75$; $P < 0.0001$), and NSC, in which it had no correlation ($P = 0.10$).

Crude protein concentrations of grasses in the current study are comparable to values reported by Allen et al. (2013) which ranged from 167 to 253 g kg^{-1}; however, PRG values were slightly lower than previously reported (202 to 253 g kg^{-1}). Marley et al. (2014) reported CP values for PRG that were slightly lower, with a mean of 157 g kg^{-1}. Smit et al. (2006) and Riberiro Filho et al. (2019) reported values of 133 to 203 g kg^{-1} for multiple PRG cultivars, which were similar to the current study results. Values for OG were similar to those reported (127 to 165 g kg^{-1}) by Marten and Hovin (1980); however they reported lower values for TF (129 to 155 k kg^{-1}). Differences in CP can be attributed to many factors, including geographical location, soil type, fertility, weather events, and grazing and harvest management.

Neutral Detergent Fiber

Grasses in year 1 contained more NDF (559 g kg^{-1}) compared to year 2 (523 k kg-1; $P < 0.0001$), and SP plots contained more NDF (548 g kg^{-1}) compared to GR plots (533 g kg^{-1}; $P = 0.0013$; Table 4). Adding clover decreased NDF (523 g kg^{-1}) relative to monoculture plots (559 g kg^{-1}; $P < 0.0001$). Comparing across the year by grass interaction ($P < 0.0001$; Table 4), OG contained the highest amount NDF in both years (591 to 601 g kg^{-1}). All PRG varieties were among the lowest in NDF in both years (473 to 544 g kg^{-1}).

In the first year of the current study, NDF values were higher in compared to those reported by Allen et al. (2013), which ranged from 396 to 547 g kg^{-1}. In the second year of that study, NDF values were more comparable to current results, ranging from 422 to 523 g kg^{-1}. In both years of the Allen et al. study (2013), similar patterns and trends between OG, TF, MF, and PRG were observed. However, the overall increase in

NDF values from year 1 to year 2 contradict the findings in the current studies. Smit et al. (2006) reported mean NDF values of 476 to 490 g kg^{-1} for six PRG cultivars, which are slightly lower than values reported in the current study. Current study results also agree with findings from Marley et al. (2014) who examined PRG under beef steer grazing (531 g kg^{-1}), and from Ribeiro Filho et al. (2019) who examined PRG and PRG-white clover under dairy cow grazing (507 to 575 g kg^{-1}). Additionally, current results are consistent with findings from Marten et al. (1987) who suggested most CSG contain between 500 and 600 g kg^{-1} NDF. Differences in NDF can be attributed to many factors, but are primarily influenced by maturity at the time of grazing and harvest. There are no recommendations for the inclusion of NDF in the diet of horses and beef cattle.

Calcium and Phosphorous

Minimal differences in calcium were observed (Table 4). Across varieties, PRG 'Forageur' and 'Spreader IV' contained more calcium (4.8 to 5.0 g kg^{-1}) than OG (3.6 g kg^{-1}; P = 0.0023). Across varieties, MF contained less phosphorous (4.1 g kg^{-1}) than all other grasses (4.3 to 4.5 g kg^{-1}; P < 0.0001). Comparing across the year by grass interaction (P = 0.0154; Table 4), OG, PRG 'Forageur' and PRG 'Spreader IV' contained more phosphorous (4.5 g kg^{-1}) in year 2 compared to the same varieties in year 1, along with MF in both years (4.0 to 4.2 g kg^{-1}). Comparing across the year by grass interaction, nearly half of the grasses had an inverted calcium to phosphorous ratio. Calcium was strongly correlated to horse DE (R^2 = 0.81; P < 0.0001).

Differences in the Ca:P can be attributed to many factors, including geographical location, soil type, fertility, weather events, and grazing and harvest management. Both DeBoer et al. (2017) and Jaqueth et al. (Jaqueth et al., 2019) also reported inverted and low calcium to phosphorous ratios in annual warm-season grasses, and cool-season turfgrasses, respectively. While intake of both calcium and phosphorous is important, the ratio between the two is equally important. For most classes of horses, the NRC (2007) recommends a ration between 1:1 and 6:1; however, if the ratio is below 1:1, supplementation of calcium is necessary to increase mineral availability for bone metabolism (Caple et al., 1982; NRC, 2007).

Nonstructural Carbohydrates

Across cultivar, PRG 'Remington' contained the greatest amount of NSC (147 g kg^{-1}) while OG contained the least (103 g kg^{-1}; $P < 0.0001$; Table 4). All other grasses contained moderate amounts of NSC (128 to 136 g kg^{-1}). The average NSC value was greater in year 2 (140 g kg^{-1}) compared to year 1 (120 g kg^{-1}; P < 0.0001) and overall, clover presence increased NSC from 127 g kg^{-1} to 132 g kg^{-1} ($P = 0.0176$). Comparing across the year by grass interaction ($P = 0.0015$; Table 4), PRG 'Remington' in year 2 contained more NSC (162 g kg^{-1}) compared to all other grasses except PRG 'Spreader IV' and MF in year 2. Year 1 OG contained the least (101 g kg^{-1}) compared to all other grasses. In year 1, PRG 'Forageur' ranked moderately low (128 g kg^{-1}) and in year 2, it ranked moderately high (143 g kg^{-1}).

Allen et al. (2013) reported similar NSC values (83 to 169 g kg^{-1}). Perennial ryegrass was within a group of grasses (Kentucky bluegrass [*Poa pratensis* L.], MF, and PRG) with greater NSC (104 to 169 g kg^{-1}) compared to meadow bromegrass (*Bromus biebersteinii* Roem. & Schult), OG, and reed canarygrass (*Phalaris arundinacea* L.; 63 to 95 g kg^{-1}). Catalano et al. (2019) also found that WC had more NSC compared to WC-CSG mixtures. The reported values were consistent with current results (110 to 124 g kg^{-1}

and 115 to 145 g kg^{-1} for WC-CSG and WC, respectively). Perennial ryegrass is notoriously higher in NSC compared to other CSG. This is perhaps why it is a sought after species for beef cattle pastures. Although a higher NSC content may be desirable to growing livestock, values too high may be detrimental for some horses. Frank (2009) suggested a total diet ≤12% NSC for horses diagnosed with EMS and Borgia et al. (2009) recommended hay containing ≤10% NSC for horses affected by PSSM.

Equine Digestible Energy

Overall, OG was lowest in equine DE (2.17 Mcal/kg) whereas all three PRG varieties were highest (2.35 to 2.38 Mcal/kg; $P < 0.0001$). All grasses had more DE in year 2 (2.38 Mcal/kg) compared to year 1 (2.27 Mcal/kg; $P < 0.0001$). The addition of white clover increased average equine DE from 2.23 to 2.38 Mcal/kg. Comparing across the year by grass interaction ($P < 0.0001$; Table 4), PRG varieties in year 2 were highest in DE (2.41 to 2.46 Mcal/kg). Year 1 PRG varieties were similar to MF and TF equine DE values in both years (2.25 to 2.31 Mcal/kg), and OG in both years was among the lowest (2.16 to 2.18 Mcal/kg).

The average 500-kg adult horse in light work requires 19.98 Mcal d^{-1} (NRC, 2007). If a horse consumed 2.0% of their bodyweight (BW) on a dry matter (DM) basis each day, the reported DE values would meet or exceed the horse's needs. Equine DE is not commonly reported for perennial CSG; however, it has been discussed for annual CSG (Grev et al., 2017), annual warm season grasses (DeBoer et al., 2017), and legumes under horse grazing (Catalano et al., 2019).

Cattle Metabolizable Energy

All PRG varieties were among the highest of cattle ME (2.45 to 2.52 Mcal/kg), and 'Spreader IV' was highest compared to all other grasses except PR 'Remington'. Orchardgrass contained the lowest cattle ME (2.37 Mcal/kg). Unlike equine DE, clover presence did not affect cattle ME ($P = 0.4537$). Year 1 grasses were higher in cattle ME (2.47 Mcal/kg) compared to year 2 (2.43 Mcal/kg; $P < 0.0001$). Across the year by grass interaction ($P = 0.0025$; Table 4), PRG 'Forageur' was moderately ranked in both year 1 (2.47 Mcal/kg) and year 2 (2.44 Mcal/kg).

Neutral Detergent Fiber Digestibility

All PRG varieties were highest in NDFD (86.7 to 88.7%; $P < 0.0001$) compared to other grasses. Similar to trends across previous nutritive values, OG had the least amount of NDFD (82.8%) compared to all other grasses except MF (84.2%) which was also similar to TF (85.2%). Grasses in year 2 had reduced NDFD (84.0%) compared to year 1 (87.7%; $P < 0.0001$). Comparing across the year by grass interaction ($P < 0.0001$; Table 4), PRG varieties in year 1 had the highest NDFD (88.6 to 91.6%) and OG and TF in year 2 had the lowest (79.9 to 82.3%). Although PRG 'Forageur' was among the highest in NDFD in year 1, it was among the lowest in year 2 (85.1%).

Neutral detergent fiber digestibility values observed in the current study were higher compared to results reported by Allen et al. (2013), which ranged from 458 to 901 g kg^{-1} for all CSG species in the study, and from 527 to 901 g kg^{-1} for PRG. Solomon et al. (2014) reported similar NDFD values for PRG, averaging 777 g kg^{-1} for diploid cultivars and 807 g kg^{-1} for tetraploid cultivars.

Livestock preference

Preference was separated by location due to the difference in livestock species grazing each experiment. The final model for horse preference included year, grass, clover, the year by grass interaction, and maturity and height as covariates ($P \leq 0.0149$). The final model for beef cow preference included year, grass, and the year by grass interaction ($P \leq 0.0009$). Both horses and beef cows readily consumed all available forage (≥ 62% removal).

Maturity and height had significant effects as covariates in the horse preference model (P ≤ 0.01); however, yield did not ($P = 0.0984$). Horses preferred grass mixtures with clover (86% removal) compared to monoculture grasses (82% removal; $P = 0.015$). Comparing across the year by grass interaction ($P < 0.0001$; Table 5), in both years, OG was among the least preferred grasses (≤ 84% removal), although MF had the greatest reduction in preference from year 1 to year 2. More differences between varieties were observed in year 2 compared to year 1, although it is uncertain why this occurred.

Maturity, height, and yield did not affect beef cow preference as covariates ($P \geq 0.0871$), nor did clover presence as a main effect (P = 0.1906). Overall, all three PRG cultivars were among the most preferred grass (75 to 78% removal) compared to MF (66% removal), OG (67% removal), and TF (68% removal). Comparing across both year and grass ($P = 0.0002$; Table 5), PRG 'Remington' in year 1 was the most preferred forage (88% removal), whereas it was among the least preferred in year 2 (62% removal). 'Forageur' was consistently moderately ranked among all grasses.

Previous work has evaluated CSG preference under both horse and cattle grazing. Allen et al. (2013) reported a range in preference when PRG and other perennial CSG were grazed by horses (46 to 95% removal). Orchardgrass was less preferred than PRG, while TF and MF ranked similarly to PRG (Allen et al., 2013). Allen et al. (2013) also reported observations of coarse leaf edges in TF, and hypothesized that this could have negatively affected horse selection. Additionally, Archer (1978, 1980), Hunt and Hay (1990), and Olson et al. (2011) reported moderate to high preference of PRG compared to other CSG under horse grazing. Casler and van Santen (2001) found that Holstein cows and heifers grazing MF and TF preferred MF.

Across both livestock and treatments, preference was negatively correlated with height ($R^2 = -0.51$; $P < 0.0001$), maturity ($R^2 = -0.49$; $P < 0.0001$) and NDF ($R^2 = -0.16$; $P = 0.0254$). It was positively correlated with CP ($R^2 = 0.34$; $P < 0.0001$), equine DE ($R^2 = 0.37$; $P = 0.0002$), and cattle ME ($R^2 = 0.43$; $P < 0.0001$). There was no correlation between preference and NSC ($P = 0.7147$).

McCann and Hoveland (1991) also reported a negative correlation between preference and maturity, whereas Allen et al. (2013) did not find a correlation between the two parameters. Allen et al. (2013) found a positive correlation between NSC and preference; however, did not find a correlation between preference and CP or maturity. Reid et al. (1967), Longland and Byrd (2006), and Smit et al. (2006) also reported a positive relationships between preference and carbohydrates; however Catalano et al. (2019) reported a negative correlation between NSC and preference when horsed grazed legumes. Previous research has also reported positive relationships between preference and eqine DE (Catalano et al., 2019) and protein content (Fontenot and Blaser, 1965; Catalano et al., 2019), and a negative relationship between preference and height (Fleurance et al., 2010; Catalano et al., 2019), maturity (Burton et al., 1956; McCann and

Hoveland, 1991; Catalano et al., 2019), and fiber content (Fontenot and Blaser, 1965; Smit et al., 2006; Allen et al., 2013). Many factors will affect animal preference, including available species, agronomic management, geographic location, and weather conditions (Marten, 1978). These results point to the multifaceted and complex relationship between forage factors and livestock preference. However, selecting forages with greater preference, and similar preference if a mixture, should lead to more efficient utilization of pasture species.

Conclusions

The primary objective of this research was to determine if the newly developed winter hardy cultivar of PRG ('Forageur') preformed similarly under livestock grazing to other commonly used perennial CSG. 'Forageur' had similar yields in year 1, but lower yields in year 2 compared to other CSG under livestock grazing. Although the maturity of 'Forageur' was similar between grazing intervals, it tended to be shorter in stature. Overall, forage nutritive value of 'Forageur' was similar to other cool-season grasses. Specifically, 'Forageur' was similar or higher in CP, tended to be lower in NDF, similar in Ca and P, moderate in NSC concentration, higher in equine DE and cattle ME, and lower in NDFD. Both horses and beef cattle readily consumed all forage options; however, OG tended to be less preferred compared to other grasses. This is among the first studies in the Midwest to evaluate new PRG cultivars under both horse and cattle grazing. This research confirms that 'Forageur' is viable PRG cultivar to use in upper Midwest livestock pastures.

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CHAPTER THREE: ESTIMATION OF ACTUAL AND IDEAL BODYWEIGHT USING MORPHOMETRIC MEASUREMENTS OF MINIATURE, SADDLE-TYPE, AND THOROUGHBRED HORSES

Summary: Adding breed type, height, and neck circumference to body length and girth circumference improves bodyweight (BW) estimation in different breeds of horses; however, equations have not been developed for all breed types. The objectives were to develop BW estimation equations for Miniature, saddle-type, and Thoroughbred horses using morphometric measurements. Measurements were collected on adult (≥ 3 yr, non-pregnant) saddle-type (n=209), adult (n=249) and juvenile (< 3 yr, n=61) Miniatures, and adult Thoroughbreds (n=100). Personnel determined body condition score (BCS), measured withers height and girth circumference at the third thoracic vertebra, body length from the point of the shoulder to the point of the buttock and to a line perpendicular to the point of the buttock, and neck circumference at the midway point between the poll and withers. Each horse was weighed using a livestock scale. Bodyweight estimations equations were developed using linear regression modeling and log transformation. Mean (±SD) BCS was 6.1 (±0.8), 5.4 (±0.6), 6.0 (±1.0) and 5.0 (±0.6) for adult and juvenile Miniatures, saddle-type, and Thoroughbreds, respectively. Bodyweight estimation equations developed through the current research were within 4% of the scale BW, and offered improvements over previous BW estimation equations and weight tapes, which were off by 5 to 25%. Owner estimated BW was within 8 to 15% of scale BW. Morphometric measurements were successfully used to develop BW equations for Miniature, saddle-type and Thoroughbred horses. The equations will assist owners and professionals with managing horse BW and will be added to the Healthy Horse app.

Introduction

It has been well documented that there is a growing population of overweight and obese horses, both in the United States and abroad (Donaldson et al., 2004; Carter et al., 2009; Brooks et al., 2010; Harker et al., 2011; Thatcher et al., 2012; Martinson et al., 2014; Catalano et al., 2016). There is not one specific reason why this change is occurring, although predominant factors include lack of owner awareness and willingness to implement and follow through with bodyweight (BW) loss programs (Morrison et al., 2017). Other factors include a management shift away from allowing natural BW fluctuations based on changing seasons and feed quality shifts towards plentiful, high quality feed availability year-round (Morrison et al., 2017). This lends itself to a steady state of elevated body condition and BW [9].

Adding to this problem is that owner perception of horse BW and body condition can be inaccurate. Wyse et al. (2008) reported an obesity rate of 45% in a group of horses used for recreational riding. Within a subset of horses considered obese by researchers, 37% of owners reported the horse was not overweight. Additionally, of the subset of horses not considered overweight or obese by researchers, 28% of owners considered the horse overweight. Similar results were observed when draft and warmblood horse owners were asked to guess the BW of their horse. On average, owners were 52 kg off the actual BW, with a roughly equal amount of over- and under-estimation (Catalano et al., 2016). Morrison et al. (Morrison et al., 2017) assessed owner ability to determine overweight or obese horses from photographs, with only 11% of respondents correctly identifying an overweight or obese horse. Additionally, there was no difference between professionals (managers and/or trainers) and non-professionals (owners) in their ability to identify an overweight horse (Morrison et al., 2017). The inability of horse owners and professionals to accurately assess BW is likely contributing to the increase in obesity and warrants development of educational materials and tools that can assist with horse BW management.

Beyond horse owner and manager awareness of excessive BW is an apparent preference for over-conditioned animals. Society appears to prefer an over-conditioned animal to one that is under-conditioned (Owers and Chubbock, 2013). Previous research (Catalano et al., 2016) discussed a possible show ring preference in which exhibitors were rewarded for having horses with full, smooth and rounded hindquarters, which are often seen in overweight animals. In one study, an overweight pony with a 6.5 body condition score (BCS) was rated as “about right” for showing (Morrison et al., 2017). This preference may be due to a lack of awareness that excessive BW contributes to insulin resistance (Geor and Harris, 2009), Equine Metabolic Syndrome (EMS) (Geor and Harris, 2009), laminitis (Geor and Harris, 2009), poor thermoregulation (Webb et al., 1990), and decreased athletic ability (Thorton et al., 1987).

Some breeds are known to be more predisposed to BW gain, including ponies and Morgans (Norton et al., 2018). It has been documented that pony breeds are at an increased risk for obesity (Giles et al., 2014) and insulin resistance (Rijnen and van der Kolk, 2003), and have a higher average BCS compared to light breeds of horses (Pratt-Phillips et al., 2010). In the same study (Pratt-Phillips et al., 2010), Morgan horses also had a higher average BCS compared to Standardbreds and Thoroughbreds. Norton et al. (Norton et al., 2018) determined several traits associated with EMS were moderate to highly heritable in both ponies and Morgan horses. These findings emphasize that these breed-types require additional management and care to prevent excessive BW gain and related disorders. By emphasizing the management and tracking of BW, horse owners can minimize potential health risks.

Using a scale to determine exact BW is ideal; however, most horse owners do not have access to a livestock scale. Weight tapes are readily available; however, their accuracy has been questioned (Ellis and Hollands, 2002; Wagner and Tyler, 2011; Martinson et al., 2014; Catalano et al., 2016). Previous research has demonstrated that accuracy of BW estimation using heart girth and body length (Hall, 1971) is improved by including breed type, height and neck circumference (Martinson et al., 2014; Catalano et al., 2016). However, equations using these measurements have not been developed for Miniature, saddle-type, or Thoroughbred horses. Therefore, the objective of this study was to develop BW estimation equations using morphometric measurements for Miniature, saddle-type, and Thoroughbred horses.

Materials and Methods

All experimental procedures were conducted according to those approved by the University of Minnesota Institutional Animal Care and Use Committee (1409-31861A).

Morphometric Measurements and Demographics

Morphometric measurements were taken on Miniature horses (n = 323) at the 2016 American Miniature Horse Registry (AMHR) Nationals in Tulsa, OK, and on Thoroughbreds at the 2014 Fall Harvest Horse Show in St. Paul, MN (n = 12) and the 2017 New Vocations Charity Thoroughbred Show in Lexington, KY (n = 88) for a total of 100 Thoroughbreds. Saddle horses were measured at the 2015 Friends of the American Saddle Horse (FASH) spring show in St. Paul, MN (n = 56), at two private farms in New

Prague, MN (n = 15) and Hugo, MN (n = 33), and across four competitive trail riding events in Alabama in 2015 (n = 110). A total of 214 saddle-type horses were measured.

At all horse shows, data were collected on adult horses that were ≥ 3 years old and non-pregnant. At the AMHR nationals, there was a large number of Miniature horses under the age of 3 years. As a result, researchers measured two different subsets of Miniature horses, ≥ 3 years (adult, n = 261) and < 3 years (juvenile, n = 62). On all equines, a similar group of trained personnel collected the following measurements: BCS on a scale of 1 (poor) to 9 (extremely fat; (Henneke et al., 1983)), height at the third thoracic vertebra (Carter et al., 2009), neck circumference at the midway point between the poll and the third thoracic vertebra (Carter et al., 2009), girth circumference at the third thoracic vertebra (Martinson et al., 2014), body length from the point of the shoulder to the mid-point of the buttock (BL wrap) (Wagner and Tyler, 2011), and body length from the point of the shoulder to a line perpendicular to the point of the buttock (BL straight) ((Martinson et al., 2014; Figure 3).

The team assessing BCS used consensus to determine the horse's final score. Body length wrap was collected for use with the Hall equation (Hall, 1971), while BL straight was collected in order to develop actual and ideal BW equations similar to other breed types (Martinson et al., 2014; Catalano et al., 2016). Additionally, each horse was weighed on a calibrated, portable livestock scale (Weigh-Tronix, Fairmount, MN, PS2000). An estimation of BW using a weight-tape, and owner-reported age, gender, breed, coat color, and discipline were also recorded. At the AMHR Nationals, owner-reported official show height was also recorded. Owners (or handlers) were also asked to estimate the horse's BW before weighing the horses on the scale, except at the four competitive trail riding events in Alabama.

Statistical Analysis and Estimation of Actual and Ideal BW

All statistical analyses were conducted using SAS (Version 9.4; Cary, NC). Summary statistics (± standard deviation [SD]) were calculated for age, BCS, height, neck circumference, girth circumference, BL wrap and BL straight. Horses were grouped by age and breed-type (adult Miniature, juvenile Miniature, Thoroughbred, or saddle-type) and groupings were confirmed using multivariate analysis of variance (MANOVA or multivariate ANOVA; (Martinson et al., 2014; Catalano et al., 2016)). All individuals within a breed-type and age group were used to develop linear models to predict BW using height, girth circumference, BL straight and neck circumference. Models based on a log transformation were also considered, and the best model was selected based on model diagnostics including root mean square error (rMSE) and the Aikeke information criterion (AIC). Leave-one-out cross-validation was used to ensure the data were not overfit. Results were compared to previously developed BW equations (Hall, 1971; Bruce et al., 2010). Only horses in each breed-type and age group with a BCS of 5 were used to develop ideal BW equations as these individuals were assumed to have an ideal BW (Harker et al., 2011; Martinson et al., 2014; Catalano et al., 2016). Only measurements not affected by adiposity were included in the ideal BW model and included height and BL straight. The best model was selected based on model diagnostics and AIC.

Other parameters examined included correlations between owner-guessed BW, scale BW and BCS, as well as the difference between BL wrap and BL straight with

BCS. The average difference between ideal and scale BW was calculated for each BCS within a breed-type and age group. For Miniature horses, the difference between height at the third thoracic vertebra and official show height were calculated, and correlations between these differences were analyzed.

Results and Discussion

Determination of Breed-Types and Age Categories

Differences between breed-types were confirmed using multivariate ANOVA analysis ($P < 0.001$, [6,7]). Model fit for Miniature horses was improved by creating separate equations for adult and juvenile horses. Owner reported saddle-type horse breeds included American Saddlebred, Morgan, Tennessee Walking Horse, Gaited Paint Horse, Kentucky Mountain Gaited Horse, Missouri Fox Trotter, Paso Fino, Racking Horse, Rocky Mountain Horse, Saddle Horse and Spotted Saddle Horse. Based on model fit, it was determined that using one equation for all saddle type-breeds was appropriate.

Demographics and Morphometric Measurements

Adult Miniature horses were comprised of 51% geldings, 36% mares and 13% stallions. Juvenile Miniature horses were comprised of 49% colts, 45% fillies and 6% geldings. It was observed that the AMHR Nationals functioned as a marketing and sales opportunity for Miniature horses and was advertised as a "National Show", which may help explain the increased prevalence of stallions compared to past research where stallions comprised <1% of measured horses [6,7]. Thoroughbreds were mostly geldings (81%), followed by mares (17%), then stallions (2%). Saddle-type horses were also predominately geldings (72%) compared to mares (28%). The increased prevalence of Thoroughbred and saddle-type geldings may be due to perceived handling differences between geldings, mares, and stallions. The preference for geldings was also observed at a draft horse show and shows where predominantly Warmblood breeds were exhibited (Catalano et al., 2016). In contrast, AMHR Nationals exhibitors shared that handling miniature horse stallions was similar to handling geldings, and the research team observed that the ability handle stallions was primarily based on their physical size. This may help explain the increased number of stallions exhibited at the AMHR Nationals.

Equine measurements are presented in Table 6. Adult Miniature horses had an average BCS of 6.1 ± 0.8, which was greater compared to juvenile Miniature horses ($P < 0.001$, 5.4 ± 0.6). No Miniature horses were considered underweight (BCS ≤ 3), while 30% of adult Miniature horses and 2% of juvenile Miniature horses were considered overweight (BCS ≥ 7). Thoroughbreds had an average BCS of 5.0 ± 0.6 while saddle-type horses had an average BCS of 6.0 ± 1.0. No Thoroughbreds were considered to be overweight, while 1% were considered to be underweight. No saddle-type horses were considered to be underweight, whereas 36% were considered to be overweight. Across all breeds, Morgan horses had the highest average BCS (6.6 ± 0.9). These results support previous literature that indicated certain breeds and breed-types including ponies and Morgan horses are predisposed to excess adiposity (Rijnen and van der Kolk, 2003; Pratt-Phillips et al., 2010; Giles et al., 2014; Norton et al., 2018).

Of the adult breed-types, Miniature horses tended to be younger (7.3 ± 4.0 years) while Thoroughbred and saddle-type horses tended to be older with mean ages of 10.2 and 10.8, respectively. Although the New Vocations Charity Thoroughbred Show was a showcase for retired racehorses, the average age of the exhibited Thoroughbred was 10.2 years (Table 6). These results help confirm the relatively short racing career many

Thoroughbreds have and highlights the potential for a lengthy post-racing career. Furthermore, these results are consistent with previously reported average ages for draft (7 years), warmblood (10 years) (Catalano et al., 2016), and stock-type horses (12 years) (Martinson et al., 2014). The relatively younger age of both draft and Miniature horses may be connected to the disciplines of these breeds (e.g. in-hand and driving exhibitions). There was a weak negative correlation between age and BCS for juvenile Miniature and saddle-type horses ($R^2 < -0.254$, $P < 0.005$). No correlation was found between age and BCS for adult Miniature horses or Thoroughbreds ($P > 0.286$).

As expected, saddle-type horse and Thoroughbred morphometric measurements were greater when compared to Miniature horses (Table 6). Juvenile Miniature horses were the lightest breed-type (91 ± 27 kg), followed by adult Miniature horses (117 ± 21 kg), saddle-type horses (473 ± 48 kg) and Thoroughbreds (538 ± 42 kg). Miniature horses were lighter compared to pony breeds previously reported, while saddle-type horses were consistent with other light breeds (Martinson et al., 2014). Thoroughbred BW was similar to those previously reported [6,7].

Estimation of Actual BW

Bodyweight estimation equations were developed using girth circumference (girth), neck circumference (neck), height, and BL straight (length) from all individuals in each breed-type and age category (Miniature horses only). All models, except the Thoroughbred model, were log transformed. The following equations were developed:

$$\text{Adult Miniature horse: } [0.000231 \times \text{Girth}^{1.836} \times \text{Length}^{0.562} \times \text{Height}^{0.319} \times \text{Neck}^{0.099}]$$

$$R^2 = 0.93, \text{ rMSE} = 4.8\%$$

$$\text{Juvenile Miniature horse: } [0.000153 \times \text{Girth}^{1.637} \times \text{Length}^{0.438} \times \text{Height}^{0.702} \times \text{Neck}^{0.149}]$$

$R^2 = 0.97$, rMSE = 5.4%

Saddle-type: $[0.000642 \times \text{Girth}^{1.600} \times \text{Length}^{0.5449} \times \text{Height}^{0.283} \times \text{Neck}^{0.222}]$

$R^2 = 0.84$, rMSE = 4.9%

Thoroughbred: $[((4.860 \times \text{Girth}) + (1.649 \times \text{Neck}) + (1.522 \times \text{Length})) - 813.947]$

$R^2 = 0.76$, rMSE = 3.9%

For Miniature horses (both adult and juvenile), estimating BW using a weight tape was off by an average of 14%, the Bruce equation (Bruce et al., 2010) was off by 5%, and the Hall equation (Hall, 1971) was off by 6%. For both saddle-type horses and Thoroughbreds, estimating BW using a weight tape was on average 5% off and the Hall equation was 25% off. The BW equations developed through this research were within 4% of the scale BW for all breed-types and ages. The use of additional morphometric measurements and breed-type have also been shown to improve the accuracy of estimating BW in other equine breed-types [6,7].

Estimation of Ideal BW

Ideal BW was estimated using BL straight and height in each breed-type and age category (Miniature horse only). Similar to the actual BW estimation equations, all models, except the Thoroughbred model, were log transformed. The following equations were developed:

Adult Miniature horse: $[0.00115 \times \text{Length}^{0.786} \times \text{Height}^{1.741}]$

$R^2 = 0.87$, rMSE = 8.0%

Juvenile Miniature horse: $[0.00005492 \times \text{Length}^{1.919} \times \text{Height}^{1.261}]$

$R^2 = 0.92$, rMSE = 9.8%

Saddle-type: $[0.00279 \times \text{Length}^{1.370} \times \text{Height}^{1.014}]$

$$R^2 = 0.59, \text{rMSE} = 7.8\%$$

$$\text{Thoroughbred: } [(1.913 \times \text{Length}) + (4.251 \times \text{Height}) - 487.900]$$

$$R^2 = 0.38, \text{rMSE} = 6.2\%$$

Differences between ideal and scale BW per BCS averaged 8, 3, 15, and 26 kg for adult Miniature, juvenile Miniature, saddle-type, and Thoroughbred horses, respectively (Table 2). Similar to past research [22], the amount of BW increase or decrease per BCS also depended on equine breed-type. Previous research estimated a 16 to 20 kg BW gain for each unit increase in BCS for mature horses (Huesner, 1993). More recently, Martinson et al. (Martinson et al., 2014) and Catalano et al. (Catalano et al., 2016) determined the average difference between BCS for Arabians, ponies, stock horses, draft, and warmblood horses was 15, 10, 17, 39, and 17 kg, respectively. This translated to 2.8 to 4.6% of BW (Martinson et al., 2014; Catalano et al., 2016). In the present study, the difference between each BCS score averaged 8 (6.8%), 3 (3.3%), 15 (3.2%), and 26 kg (4.9%) for adult Miniature, juvenile Miniature, saddle-type, and Thoroughbred horses (Table 2). These numbers are similar to previous research (Martinson et al., 2014; Catalano et al., 2016); however, the literature is limited on the evaluation of the impact of BW on BCS of young growing horses, highlighting a need for future research.

The difference between the actual scale weight and estimated ideal BW increased as BCS moved above a score of 5 and decreased as BCS decreased (Table 7). Thoroughbreds with a BCS of 3 were estimated to be 12% underweight, while adult Miniature horses with a BCS of 8 were estimated to be 16% overweight. Although this data appears to follow a relatively linear relationship, it has been suggested the amount of BW change needed to observe one unit change in BCS may not be linear, and may differ between increasing and decreasing BCS (Dugdale et al., 2012). Further research into differences between BW loss and BW gain are warranted. However, using ideal BW estimations can give owners and managers a goal BW to use in nutritional and BW management.

Owner Estimated BW

Owners were somewhat able to guess equine BW (adult Miniatures, $R^2 = 0.65$; juvenile Miniatures, $R^2 = 0.83$; saddle-type, $R^2 = 0.64$; Thoroughbred, $R^2 = 0.45$; $P < 0.01$). On average, adult and juvenile Miniature horse owners were within 15% (16 kg) of correctly guessing scale BW. On average, saddle-type horse and Thoroughbred owners were within 8% of the scale BW. This translates to roughly 38 and 43 kg, respectively.

Owner estimation of horse BW has not been examined in depth except for within draft and warmblood breeds (Catalano et al., 2016). On average, draft and warmblood horse owners were off by 7% (52 kg) when estimating BW. The ability to accurately estimate BW is critical since many management aspects are based on BW, including dosage of anthelmintics and other medical treatments, and ration formulations. If owners and managers are unable to accurately estimate BW, it may lead to incorrect medical dosing or excessive or insufficient energy and other dietary nutrients in the ration. In addition to BW, researchers have investigated an owner's ability to estimate BCS (Jensen et al., 2016; Morrison et al., 2017). In a study assessing owner ability to determine excess adiposity on horses from a photograph, only 11% of survey respondents could correctly identify an overweight or an obese horse [8]. A study assessing BCS in Icelandic horses found that 42% of owners underestimated BCS [25].

Differences between Actual Height and Show Height

At the AMHR show, Miniature horses were placed into categories or classes based on height at the base of the last mane hair. The maximum height allowed for breed registration in the AMHR is 86.4 cm for the “under” category (class A), and 96.5 cm for the “over” category (class B). Most Miniature horse owners shared they felt it was more advantageous to have the tallest equine in each category or class. Therefore, the research team observed that the term “last mane hair” was used loosely as owner’s commonly used body clipping and hair dye as a method to establish the “last mane hairs”. Owner-reported show height, or the measurement recorded as the AMHR official height, was recorded at the time of data collection. The average height difference between show height and withers height (third thoracic vertebra) measured during the research trial was 5 cm. Although the research team anticipated a difference between withers height and show height, the interpretation of “last mane hair” was surprising.

Healthy Horse App

To remove the technical barriers that may inhibit some horse owners and managers from using the new BW prediction equations, all equations have been added to the fee-based “Healthy Horse” app. This app is available for use with Android (Google Play store) and Apple (iTunes store) operating systems. To use the app, owners and managers select a breed type and enter the height, girth circumference, BL straight, and neck circumference, and the app calculates the horse’s estimated and ideal BW. Imperial or metric units can be used, and the app is offered in both English and Spanish. Bodyweight estimation equations used in the app for Arabians, ponies, and stock horses were developed by Martinson et al. (Martinson et al., 2014), while equations for draft and warmbloods were developed by Catalano et al. (Catalano et al., 2016).

Conclusions

All horses had a mean BCS between 5.0 and 6.1, while mean BW ranged from 91 to 538 kg. Separating horses into breed-types and adding height and neck circumference, along with BL straight and girth circumference, resulted in improved BW estimation equations. Height and BL straight were used to develop ideal BW equations, and differences between ideal and scale BW per BCS averaged 8, 2, 15, and 26 kg for adult Miniature, juvenile Miniature, saddle-type, and Thoroughbred horses, respectively. Saddle-type horse and Thoroughbred owners were within 8% of horse scale BW, while Miniature horse owners were within 15% of the scale BW. A difference was observed between withers height and show height for Miniature horses; however, it is unclear if this will translate to any lasting impacts on the breed type.

Development of more accurate equations to estimate actual and ideal BW, specifically for adult and juvenile Miniature, saddle-type, and Thoroughbred horses, should help owners and professionals better manage these breed types. Owners and professionals can use the new equations, in conjunction with BCS, to formulate rations with appropriate caloric intake, accurately dispense medication, track changes in BW over time, and establish BW goals. To remove technical barriers that may inhibit use, all BW equations were added to the Healthy Horse app.

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CHAPTER FOUR: THE EFFECT OF GOLDFISH (*CARASSIUS AURATUS*) ON WATER QUALITY IN HORSE STOCK TANKS

Summary: Goldfish (*Carassius auratus*) have been reported as a method to keep water tanks clean; however, little information exists on this approach. The objectives were to evaluate the efficacy of goldfish on maintaining water quality in tanks and to evaluate the frequency that this method is used. The first objective was completed June through October 2017 in St. Paul, MN using plastic and metal 379 L stock tanks, each with and without goldfish in a drylot that housed six adult horses. The stocking rate was 5 goldfish per tank, and all tanks were refilled when any tank reached 190 L. Daily readings of total dissolved solids (TDS) and water turbidity (NTU), and weekly samples to measure chlorophyll *a* were taken. At the end of each 28-day period, tanks were cleaned and rotated. Plastic tanks had lower TDS compared to metal tanks ($P < 0.001$); however, metal tanks had lower NTU and chlorophyll *a* ($P \leq 0.008$). Adding goldfish resulted in lower TDS ($P < 0.001$); however, there was no effect on NTU or chlorophyll *a* ($P \geq 0.097$). The second objective was completed using an online survey that was open from October 31 until December 15, 2018. Of the 672 completed surveys, 56% had not tried using goldfish in water tanks, 26% had utilized goldfish in the past, and 18% currently used goldfish. The inclusion of goldfish in water tanks did not affect all water quality parameters; however, 44% of survey respondents had tried, or were currently using, this management method.

Introduction

Water is often overlooked as one of the seven key nutrients required by horses. Water is essential for body fluid balance, digestive function, and gastrointestinal health (NRC, 2007). Daily requirements range from 21 to 29 L per day for an idle 500-kg horse on a hay-based diet, although this varies based on a number of conditions including workload and ambient air temperature (NRC, 2007). Water intake occurs directly by drinking fluid water and by consuming feed that contains a high percentage of moisture, and indirectly though metabolism of macronutrients (NRC, 2007). It is generally understood that pasture can provide horses with some water; however, when horses are on a primarily hay-based diet, the majority of their water requirements will be fulfilled through drinking water. Ensuring good water quality is paramount to maintaining overall health in the horse (Hinton, 1978; Jones, 2004; NRC, 2007; Lardy et al., 2008).

Water quality standards, which determine the suitability for consumption, vary widely (Markwick, 2007; NRC, 2007). The primary parameter for assessing water quality for livestock use is the total dissolved solids (TDS) value. Other parameters include odor, color, temperature, and turbidity. Total dissolved solids is a measure of the aggregate composition of the ions in the water (NRC, 2007). Total dissolved solids values of under 1,000 mg/L are considered safe and should pose no health risks. Values between 1,000 and 2,999 mg/L are generally safe, but may cause temporary diarrhea in unaccustomed horses. Values above 3,000 mg/L may negatively affect water intake, while water with values above 5,000 mg/L should be avoided for pregnant or lactating animals [6]. Water with TDS values above 7,000 mg/L should not be offered (Environmental Studies Board, 1972). However, TDS is generally low (< 350 mg/L) in North American water sources (NRC, 2007).

Algae are a growing problem in both naturally occurring and man-made bodies of water, including stock tanks commonly used to provide water to horses and other livestock. Of the contaminants that affect water quality, algae are some of the more visible ones and can be a signal to clean the water source. Algae are photosynthesizing organisms that make their own food by using solar energy, and under the right temperature and water conditions, can grow very rapidly and form extremely high-density populations or blooms (Sheath and Wehr, 2015). Algae blooms can turn the water green and affect odor, which may negatively affect intake rates (NRC, 2007). Blooms can also potentially be toxic and pose serious health risks to people, pets, and livestock causing gastrointestinal diseases and death [7].

To our knowledge, there is no research examining cleaning methodology for livestock water tanks, even though water quality is emphasized to be of upmost importance (Hinton, 1978; Jones, 2004). There is an anecdotal concept that common goldfish (*Carassius auratus*) can be used to help maintain water quality in livestock water tanks. In theory, goldfish eat algae and other contaminants such as dropped feed, thus maintaining good water quality. Some owners believe by including goldfish, the tank stays cleaner for a longer duration, reducing the overall labor and time spent cleaning tanks. Limited data exists to support this concept; however, one study investigating the feasibility of using common goldfish to control algae in urban drainage systems reported positive results (Scholz and Kazemi-Yazdi, 2005). Goldfish are classified as herbivores, particularly when applied to closed ponds or other water systems (Scholz and Kazemi-

Yazdi, 2005). If substantiated, this method offers a viable option for horse owners and managers to reduce labor and time spent cleaning tanks. Therefore, the objectives of this research were to evaluate the efficacy of goldfish on maintaining water quality in tanks and to evaluate the frequency this method is used in the horse industry.

Materials and Methods

Water Quality Experiment

All experimental procedures were conducted according to those approved by the University of Minnesota Institutional Animal Care and Use Committee (protocol #1704-34718A).

The study was conducted June through October 2017 in St. Paul, MN. Black polyethylene (plastic) and galvanized steel (metal) 379 L stock tanks were used both with and without goldfish. Tanks were placed along the fence line of a drylot that housed six stock-type, adult horses (526 kg ± 30, 5.5 ± 0.8 body condition score [BCS]; (Henneke et al., 1983)) overnight, and when horses were not grazing on pasture. Tanks were approximately three meters from each other and 0.3 meters away from the fence line. Prior to the start of the experiment, tanks were marked to allow determination of water volume at 38 L intervals. When any one tank was below 190 L, all tanks were refilled to 379 L. The amount of water added was used to evaluate horse preference. At the end of each period (28 days), tanks were rotated in position along the fence line. This was done to alleviate any potential horse bias due to location. One metal tank and one plastic tank were stocked with five common goldfish, purchased from local pet stores. Goldfish weight was unable to be determined in a reliable or consistent fashion; however, goldfish were consistently 3 to 4 cm long. Stocking rate was determined based on store recommendations of 76 to 95 L per goldfish. In the event of goldfish death, a replacement was added within 24 hours. Replacement goldfish were kept in a separate 379 L stock tank located immediately outside of the drylot.

Total dissolved solids and water temperature were measured daily using a handheld meter (HM Digital COM-100; HM Digital, Inc., Redondo Beach, CA). The TDS value received from the handheld meter was validated ($R^2 = 0.82$, $P < 0.001$) by submitting biweekly samples to Minnesota Valley Testing Laboratories (New Ulm, MN). Both measurements were taken at nine points within each tank using three replicates at a high, mid, and low water level. The low water level was taken at the 38 L mark each day, while the mid-point was taken at the average water level of the low and high-points. The high-water level was taken immediately below the water surface. The three replicates were taken evenly across the tank, from left to right lengthwise. Daily water turbidity was determined using a Secchi tube (Water Monitoring Equipment & Supply, Seal Harbor, ME) by placing the tube diagonally across the tank lengthwise. If water level was insufficient to fill the tube, additional water was added from the tank using a measuring cup until the tube was filled. Water was released from a valve at the bottom of the tube until a black and white disc was clearly viewed when looking directly down into the tube. Water level within the Secchi tube was recorded in centimeters and converted to natural turbidity units (NTU) using [$NTU = (cm / 244.13)^{1/-0.662}$; (Myre and Shaw, 2006)). Air temperature was recorded daily from a nearby university weather station. Plywood was used to provide shade over half of each tank, including those without goldfish, in an attempt to keep water temperature within a viable range for the goldfish and to provide protection from predators (e.g., birds). Water samples were taken on a weekly basis for chlorophyll *a* evaluation. Chlorophyll *a* was used as an indicator of algae presence, due to this pigment being produced by all major algal groups found in freshwater (Sheath and Wehr, 2015).

Horse Owner and Manager Survey

The University of Minnesota Horse Extension team administered a short survey (10 to 14 questions) regarding the use of goldfish in tanks. The target audience for the survey was horse owners and managers. The survey was advertised on the team's Facebook page, in their monthly electronic newsletter, and at in-person Extension events. The survey was available (e.g., open) from October 31 to December 15, 2018. The survey consisted of five to nine questions regarding water tank and fish use, along with five demographic questions. The first survey question asked if a participant had previously used, currently used, or had never used goldfish or other aquatic species in tanks. Based on the response to the first question, participates were asked slightly different questions; however, all participates were asked the same demographic questions. Water tank and fish use questions focused on the size of tanks used, use and stocking rate of fish, tank cleaning frequency, the respondent's thoughts on how effective fish were at maintaining tank water quality, and if research on the subject would influence management decisions.

Statistical Analysis

All analyses were conducted using SAS 9.4 (Cary, NC). Regression analysis was conducted using PROC MIXED to determine effects of goldfish and tank type on TDS, NTU, chlorophyll *a*, horse preference, and water temperature. Ambient air temperature was used as a covariate in the water temperature model. Water temperature and water level were evaluated as covariates on all other regression analyses. Tank type and inclusion or exclusion of goldfish were treated as fixed effects, while replicate was a random effect in all models. Period and the period by week interaction were evaluated as both random and fixed effects; the decision was made based on fit statistics. Other interactions (e.g. tank by goldfish and week by tank by goldfish) were also considered and removed if not significant ($P \leq 0.05$). Means are the least squared means of the MIXED procedure (± SE). The results of the handheld TDS meter and laboratory TDS values were evaluated using PROC CORR. PROC FREQ was used to create summary statistics for the horse owner and manager survey.

The final model for water temperature included water level, period, week, air temperature, and the period by week interaction in the main model, with air temperature as a covariate. The final model for TDS included tank, goldfish, week, the tank by goldfish interaction, and the tank by goldfish by week interaction as fixed effects, and water temperature and water level as covariates. Replicate, period, and the week by period interaction were random effects. The final model for NTU included tank and week as fixed effects and water temperature as a covariate. Replicate, period, and the period by week interaction were random effects. The final model for chlorophyll *a* included tank, week, and goldfish. The final model for horse water consumption (preference) included period, tank, fish, location within the dry lot, and the tank by fish interaction.

Results and Discussion

Water Quality Experiment

Water Temperature

There was no effect of water level (high, mid, low; $P = 0.092$), presence of goldfish ($P = 0.208$), or tank type ($P = 0.244$); therefore, these parameters were removed from the model. All other main effects and interactions were significant ($P < 0.001$). Average monthly water temperatures (Figure 4) were warmest in June (22.0°C ± 0.05), followed by July (21.7°C ± 0.06), August (20.4°C ± 0.05), September (19.8°C ± 0.05), and October (18.0°C ± 0.07). For every degree increase in air temperature, water temperature increased by 0.60 degrees. For every 1 L increase in water level, water temperature increased by 0.01 degrees. Although it is generally understood that darker colors will absorb UV light, and have the energy potential for heat, there was no difference between tank types, even though the metal tank appeared lighter in color compared to the plastic tank, which was black. This is likely due to the fact that water was being added to all tanks in a frequent and consistent manner, helping to maintain a more constant temperature.

The effect of air temperature on water temperature, and subsequent effects of water temperature on water quality parameters explained below, highlights the need to regulate water temperature, especially during warmer, summer months. Research using exercised horses found that they preferred a lukewarm (20°C) saline solution to cool (10°C) or warm (30°C) solutions (Butudom et al., 2004). In the current study, monthly average water temperature was between 23 and 18°C, similar to the preferred solution temperature listed above. Maintaining water temperature can be done in a number of ways, including covering part of the tank with plywood or similar materials, locating the tank under a shade structure, or by frequently adding or changing the water.

Total Dissolved Solids

All effects and interactions were significant ($P \leq 0.007$). Plastic tanks had a lower average TDS value (234 mg/L ± 3) compared to metal tanks (256 mg/L ± 3; Table 8).

Tanks containing goldfish also had a lower average TDS value (243 mg/L ± 3) compared to tanks without goldfish (247 mg/L ± 3). Within period, week one had a higher average TDS value (260 mg/L ± 5) compared to weeks 2 through 4 (237 to 242 mg/L ± 5). When considering the tank by goldfish interaction, plastic tanks with goldfish had lower TDS values compared to plastic tanks without goldfish and metal tanks with and without goldfish. For every one degree increase in water temperature, TDS increased by 0.80 mg/L, and for every 1 L increase in water level, TDS increased by 0.03 mg.

In the current study, mean TDS was always below 1,000 mg/L, the current threshold for safe livestock water (Environmental Studies Board, 1972). The tanks were emptied and thoroughly cleaned every 28 days, likely contributing to the acceptable water quality seen throughout the study. However, plastic tanks were observed to have an increased surface roughness compared to the metal tanks, which has been demonstrated to negatively impact cleaning ability (Blossey, 2003). Additionally, it is known that live goldfish produce waste. Although we hypothesized that any goldfish feces would gather at the bottom of the tank, and thus not affect the top drinking surface of the tank, it is possible that the combination of goldfish waste and the surface roughness of the plastic tanks resulted in the observed increase in TDS. Future research should focus on the effectiveness of goldfish when tanks are not cleaned or cleaned less frequently. The correlation between rising water temperature and increasing TDS should also serve as a reminder for the importance of tank cleaning, especially during the warm summer months.

Water Turbidity

The presence of goldfish and water level were not significant ($P \geq 0.683$) and were removed from the model. All other parameters in the final model were significant ($P < 0.001$). Metal tanks had a lower average NTU value (9.3 ± 1.7) compared to plastic tanks (18.4 ± 1.7; Table 8). Within each period, week one had lower NTU values (2.0 ± 2.4) compared to weeks two through four. Week four had higher NTU values (25.0 ± 2.2) compared to weeks one and two (11.5 ± 2.3), while week three (16.9 ± 2.3) was not different from weeks two or four. For every one degree increase in water temperature, NTU values increased by 0.65.

Turbidity is the measure of relative clarity of a liquid. Although little data exists on the connection between water turbidity and livestock health, Fraser et al. (Fraser et al., 1998) found positive correlations between turbidity and both livestock fecal coliform discharge and concentration levels, confirming that livestock waste has a negative impact on water quality. A clear correlation has also been observed in human health. Schwartz et al. (Schwartz et al., 1997, 2000) found a correlation between increasing water turbidity and hospitalizations of the elderly and young children in Philadelphia, even though NTU levels were very low (<0.3). In their study examining the effect of common goldfish on controlling algae in urban drainage systems, Scholz and Kazemi-Yazdi (Scholz and Kazemi-Yazdi, 2005) reported higher NTU values in ponds planted with aquatic vegetation and lower NTU values in unplanted ponds after goldfish were introduced. These results agree with the current study that goldfish did not necessarily affect water quality and that other factors such as tank type, water temperature, or vegetation likely play a key role in water quality. In the current study, the increase in NTU over time, and as the temperature increased, highlights the importance of frequent tank cleaning, especially during the warmer summer months.

Chlorophyll *a*

Tank and week effected chlorophyll *a* ($P \leq 0.020$) whereas inclusion of goldfish did not ($P = 0.109$). As a covariate, water temperature did not have an effect ($P = 0.155$); however, water temperature and chlorophyll *a* were only moderately correlated ($R^2 = 0.32$; $P = 0.001$). Period was not significant ($P = 0.140$) in the main model; however, it was included as a random effect along with the period by week interaction based on model fit statistics. Plastic tanks contained more chlorophyll *a* (13.2 $mg/m^3 \pm 2.7$) compared to metal tanks (7.1 $mg/m^3 \pm 2.7$; Table 8). Chlorophyll *a* increased steadily throughout each period. There was more chlorophyll *a* present in week four (19.71 $mg/m^3 \pm 3.54$) compared to week one (1.39 $mg/m^3 \pm 3.54$).

The differences in chlorophyll *a* observed between tanks was unexpected. The plastic tanks were made from polyethylene, which is a molded polymer. These tanks had a slight grit, or texture, compared to the metal tanks which were formed via sheet metal and had a smoother surface. On a microscopic level, the smoother finish was more hydrophobic. This may enhance cleaning ability compared to a rougher surface, which had a higher potential to hold both water droplets and contaminants (Blossey, 2003). Although all tanks were thoroughly cleaned at the end of each 28-day period, we hypothesize that small amounts of algae remained in the more textured surface of the plastic tanks and may have contributed to higher chlorophyll *a* levels. Over time, the surface roughness of polyethylene may increase as the product breaks down (Cooper et al., 1993). This may result in plastic tanks being harder to clean as they age. The water temperature range observed in the current study has been documented to support algal growth (Butterwick et al., 2004). While seasonal temperature swings can impact algal growth, the temperature differences observed in the current study were within the range that supports normal growth. This could help explain why water temperature did not have an impact on chlorophyll *a* levels in the current study. The increase in chlorophyll *a* over time highlights the importance of frequent tank cleaning.

Horse Water Consumption Preference

Water consumption was used to determine horse preferences. No model parameters had an impact on water consumption or horse preference ($P \geq 0.108$).

Horses are known to be selective grazers (Hunt and Hay, 1990); therefore, most research has focused on examining preference among grass (Allen et al., 2013; DeBoer et al., 2017; Grev et al., 2017) and legume (Catalano et al., 2019) pasture species. To the best of our knowledge, this is the first study documenting a lack of preference between tank types and the inclusion of goldfish. Horse owners and managers should feel confident that tank type (metal or plastic) and the inclusion of goldfish should not negatively affect horse water consumption or preference.

Fish Mortality

A total of 183 fish were purchased for this study; however, only 12 remained on the last day of the research project (140 days later; 5 goldfish in each tank and 2 in the replacement tank). This translated into a mortality rate of 93% for the goldfish. It is unclear why the goldfish struggled to thrive during the study. Water in the tanks was within normal ranges for pH, hardness, alkalinity, and total ammonia (NH_3/NH_4) based on testing at a local pet store. Water was tested and did not contain any measurable amount of chlorine. There was no evidence of bird predation, or goldfish injury or death from the horses. Goldfish are notoriously hardy and know to survive a wide temperature range (Ford and Beitinger, 2005). Fry et al. (Fry et al., 1942) reported lower and upper lethal temperatures of 0 and 41°C, respectively, for goldfish. Ford (Ford and Beitinger, 2005) reported similar results, with a minimum temperature of 0.3°C and maximum temperature of 43.6°C for goldfish. Water temperatures were well within these ranges for the duration of the current study. During water changes that occurred at the end of each 28-day period, goldfish were acclimated by being contained within a small bucket filled with water from the previous period. The small bucket floated in the freshly filled tank for 30 to 60 minutes to allow for temperature acclimation. The majority of fish deaths; however, were observed in the first days following a new period which corresponded to a complete water change. As a result, it is not recommended to completely change water when utilizing goldfish; however, this may inhibit an owner or manager's ability to maintain good water quality in tanks over time.

Horse Owner and Manager Survey

To the best of our knowledge, this is the first survey investigating the use of goldfish in horse or livestock water tanks. A total of 748 people responded to the survey, and 90% (n = 672) completed all questions; only completed surveys were used for analysis. Survey respondents were asked about fish use in horse water tanks, which included goldfish and other fish species. Fifty-six percent of respondents had never heard of, nor used, fish species in their horses' water tanks, 26% had utilized fish previously, and 18% currently utilized fish. When asked why respondents had not utilized fish, the primary reason was "I have never heard of this" (37%), followed by "other" (19%), "I do not think this would work" (16%), "fish survival or welfare concerns" (12%), "I have no interest in trying this" (10%), and "fish feces and/or livestock welfare concerns" (7%). When asked if research investigating this topic would influence their decision, 62% answered "yes", 13% said "no", and 25% said "maybe".

Survey participants who responded "yes, I previously have" or "yes, I currently do" to the question regarding fish species usage were asked if they kept the fish in the tanks year-round. Sixty-seven percent reported leaving fish in the tanks year-round, 16% reported that they brought the fish inside for the winter season, and 17% reported that the fish did not survive winter and were replaced annually. These participants were also asked if they thought the use of fish species helped keep tanks clean and reduced cleaning efforts. Sixty-one percent believed fish had a positive effect on water quality, 15% did not think that fish were effective, and 24% were uncertain. Of those that currently use fish, 75% found the fish effective at maintaining water quality, 9% did not find them effective, and 16% were uncertain. Within those that had previously used fish, but were not currently using them, 52% believed fish were effective at positively impacting water quality, 19% did not find them effective, and 29% were uncertain. Participants who had previously used fish were asked why they stopped using them. Twenty-two percent stopped because they did not feel it worked or reduced the time spent cleaning, 43% stopped because the fish died, and 35% stopped for other reasons. The most common "other" reason beyond effectiveness and fish survival was switching from stock tanks to automatic waterers.

All participants were asked how frequently they clean their horses' water tanks. Overall, 27% clean tanks once per week. Three percent of respondents clean more than once per week, 16% clean tanks two times per month, 20% clean once per month, 17% clean 3 to 4 times per year, 3% clean ≤ 3 times per year, 12% never cleaned their horses' tanks, and 3% responded with "other". Of those who never cleaned tanks, 86% currently use, or have previously used, fish species. Of those that clean tanks more than once per week, 100% of respondents had never used fish species.

The most frequently used tank size was a 379 L tank (57%), followed by 190 L (18%) and 758 L (15%). Other tank sizes ranged from 19 L buckets to 56,850 L tanks. Those that use, or had previously used, fish species reported tank sizes that ranged between 190 and 3,790 L. Common goldfish were the predominant fish species used (88%); however, respondents also reported using Koi (*Cyprinus carpio haematopterus*; 2%), Minnows (*Cyprinidae* sp.; 3%), and other fish species (7%) including Catfish (*Siluriformes* sp.), Bullheads (*Ameiurus melas*), and Plecostamus (*Hypostomus Plecostomus*). Looking at 379 L tanks, the same used in the current study, 39% were stocked with 1 to 3 fish, 41% were stocked with 4 to 6 fish, 14% were stocked with 7 to 10 fish, and 6% were stocked with 11 to 15 fish. It is unclear why there is a wide range of fish stocking rates; however, this could be due to varying fish sizes, especially as fish age and grow. Participants were not asked to estimate fish size.

Fish usage by geographic location is reported in Table 9. Current or previous fish use appeared to be most prevalent in the Midwest (54%) compared to the Mid-Atlantic, New England, Rocky Mountain, and southern regions of the U.S., along with areas outside of the U.S. This may explain why University of Minnesota faculty and staff receive frequent questions surrounding this management method. The western and southwestern regions of the U.S. previously have, or currently used, an aquatic species at rates similar to the Midwest (59 and 64%, respectively). For regions outside of the Midwest, western and southwest regions, 0 to 43% of respondents had previously used, or currently used an aquatic species in their tanks.

The majority of survey respondents were from the Midwest region of the U.S. (50%), followed by the south (13%), outside of the U.S. (13%), the Mid-Atlantic region (7%), New England (6%), the west (6%), the Rocky Mountains (3%), and the southwest (2%). The majority of respondents were female (94%), and most owned or managed ≤ 5 horses (65%), followed by 6 to 10 horses (16%), ≥ 16 horses (12%), or 11 to 15 horses (7%). Most respondents had spent ≥ 16 years owning and managing horses (55%), followed by 11 to 15 years (17%), 6 to 10 years (15%), and ≤ 5 years (13%). Fifty percent of respondents were between 21 and 40 years old, 30% were between 41 to 60 years, 11% were ≥ 60 years, and 10% were ≤ 20 years old. These demographics are consistent with the overall population that owns horses in the U.S. (Martinson et al., 2006; Stowe, 2012; Mastellar et al., 2018; Heuschele et al., 2018) and are reflective of the demographics of the e-newsletter (Martinson et al., 2010) and Facebook page used to distribute the survey.

Finally, respondents were invited to include additional comments or insights they had on the subject. Twenty-five comments referenced using fish species for mosquito (*Culicidae* sp.) and/or other insect control. These respondents mentioned that even if they didn't notice a difference in algae, they felt the fish were effective at controlling insect eggs and/or larvae. Taking insect egg and larvae counts was beyond the scope of this research. However, the research team did observe noticeable amounts of mosquito and bloodworm (*Glycera* sp.) eggs, larvae, and adults on the surface and bottom of tanks that did not contain goldfish. In comparison, there was no evidence of insects in the tanks that contained goldfish. Future research should investigate the use of fish species in controlling insect larvae in water tanks. Other notable comments included concerns about fish survival after recently dewormed horses placed their muzzles in the water, and that particularly playful horses may pose a welfare or survival issue for fish.

Conclusions

This research found that goldfish do not have a positive impact on water quality parameters in horse stock tanks, with the expectation of TDS. Tanks containing goldfish had a lower average TDS value (243 mg/L ± 3) compared to tanks without goldfish (247 mg/L ± 3). However, tank type and water temperature do appear to have an impact on water quality parameters. For every one degree increase in water temperature, TDS increased by 0.80 mg/L and NTU values increased by 0.65. Plastic tanks contained more chlorophyll *a* (13.2 mg/m^3 ± 2.7) and had a higher NTU value (18.4 ± 1.7) compared to metal tanks that contained 7.1 mg/m^3 ± 2.7 chlorophyll *a* and had an NTU value of 9.3 ± 1.7. Additionally, chlorophyll *a* and NTU increased from week 1 to week 4.

In a survey investigating the use of fish species in horse water tanks, 56% had never heard of using, nor used, fish species in their horses' tanks, 26% had utilized fish previously, and 18% currently utilized fish. Common goldfish were the predominant fish species used (88%) with a stocking rate of 1 to 6 fish per 379 L tanks. Most (27%) horse owners and managers indicated they cleaned water tanks once per week; however, 12% had never cleaned their horses' tanks. Of individuals who had never cleaned their tank, 86% currently use, or had previously used, fish to maintain water quality. Of those who no longer use fish, 22% stopped because they felt fish did not work or did not reduced time spent cleaning, while 43% stopped because the fish died. The majority of 672 respondents were from the Midwest (50%), were female (94%), owned or managed ≤5 horses (65%), had spent ≥16 years owning and managing horses (55%), and were between 21 and 40 years old (50%).

Combined, these results highlight the importance of frequent tank cleaning, especially during the warmer summer months and when using plastic tanks. In the current study, goldfish mortality rate was 93%. Based on a combination of mortality and their lack of ability to maintain all water quality parameters, we cannot conclusively recommend using goldfish as a management method for maintaining water quality in water tanks. Furthermore, goldfish should not replace frequent cleaning as a way to maintain water quality in tanks.

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APPENDIX

Table 1. Test of fixed effects for yield, the grass to white clover ratio (G:C) forage nutritive values including crude protein (CP), neutral detergent fiber (NDF), calcium (Ca), phosphorous (P), NSC (nonstructural carbohydrates), equine digestible energy (equine DE), cattle metabolisable energy (cattle ME), and neutral detergent fiber digestibility at 48 hours (NDFD) for cool season grasses grown in St. Paul (2016-2017) and Grand Rapids, MN (2017-2018).

Effect	yield	G:C	CP	NDF	Ca	P	NSC*	equine DE*	cattle ME*	NDFD*
						Pr > F				
Location (L)	<0.0001	<0.0001	0.7456	0.0013	<0.0001	<0.0001	–	–	–	–
Year (Y)	<0.0001	0.0012	0.0626	<0.0001	0.7041	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Grass (G)	<0.0001	0.1316	<0.0001	<0.0001	0.0023	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Clover (C)	0.198	–	<0.0001	<0.0001	<0.0001	0.766	0.0176	<0.0001	0.4537	0.0038
L x Y	0.3107	0.0008	0.8515	<0.0001	0.0175	0.4129	–	–	–	–
L x G	0.9226	0.3532	0.0189	0.964	0.0621	0.3704	–	–	–	–
L x C	0.6883	–	<0.0001	<0.0001	0.0026	0.0196	–	–	–	–
Y x G	0.0402	0.165	<0.0001	<0.0001	0.0602	0.0154	0.0015	<0.0001	0.0025	<0.0001
Y x C	0.973	–	<0.0001	0.1122	0.7622	0.0296	0.0002	0.5091	0.0905	0.8069
G x C	0.9299	–	0.0344	0.3503	0.2471	0.0989	0.146	0.0067	0.8958	0.8699
L x Y x C	0.1837	–	0.2462	0.0506	0.663	0.8994	–	–	–	–
L x Y x G	0.2298	0.2760	0.0493	0.6862	0.4587	0.0512	–	–	–	–
L x G x C	0.5234	–	0.4467	0.1317	0.8121	0.3831	–	–	–	–
Y x G x C	0.5147	–	0.0682	0.0006	0.1205	0.5352	0.3758	0.1339	0.6646	0.6342
L x Y x G x C	0.81	–	0.3681	0.3972	0.7111	0.8042	–	–	–	–
Maturity	0.7531	0.1225	0.0961	0.6808	0.1903	0.5383	0.2398	0.4251	0.0887	0.266

*NSC and horse DE values are for SP only and cattle ME and NDFD are for GR only, therefore location was not applicable

Figure 1A-D. Precipitation totals for St. Paul (A) and Grand Rapids (B), MN and average monthly temperature for St. Paul (C) and Grand Rapids (D), MN during the grazing season.

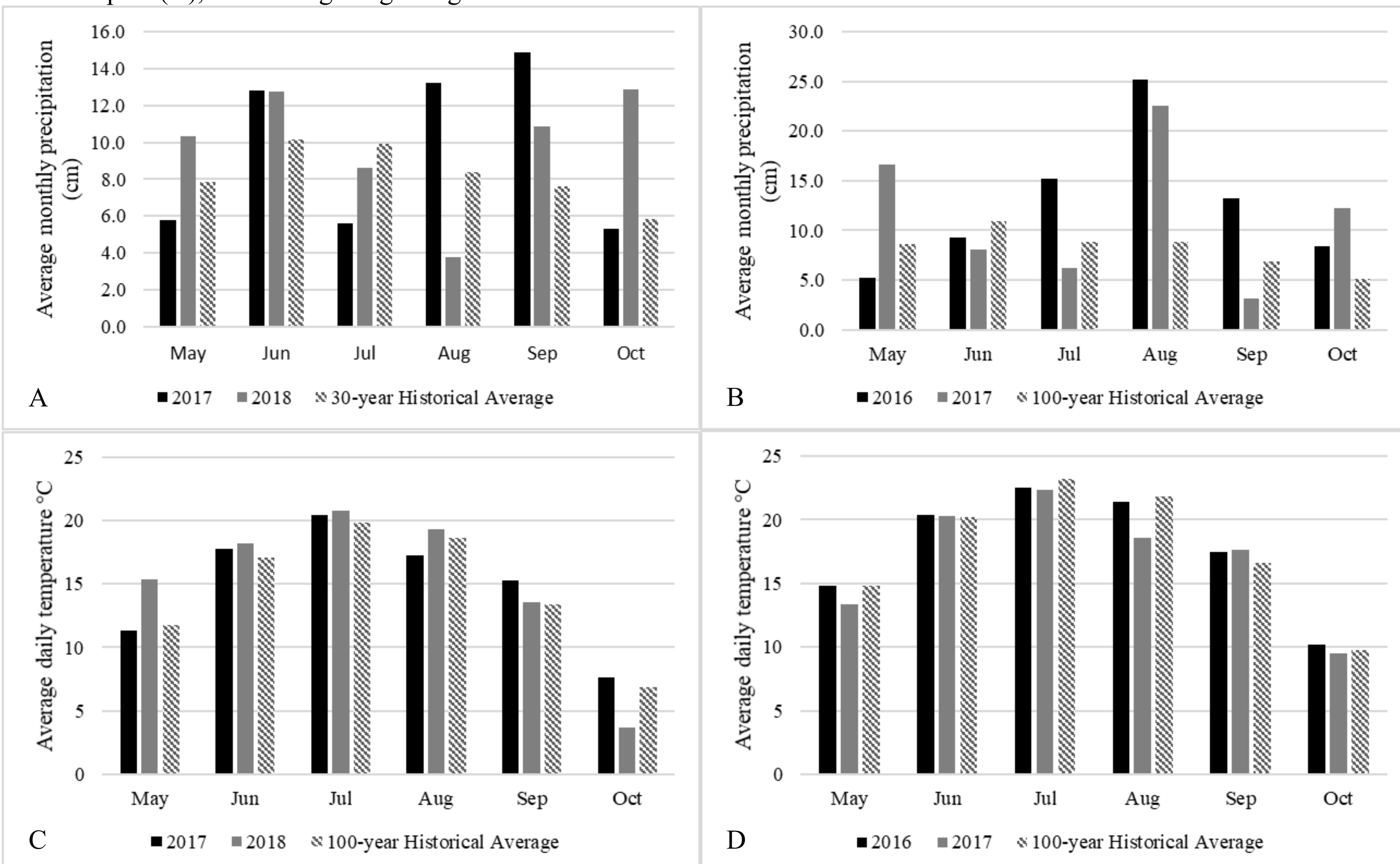

Table 2. Yield, maturity, and height of cool-season grasses grazed by livestock in St. Paul and Grand Rapids, MN.

Year by Grass		**Yield**	**Maturity**	**Height**
Year	**Grass***	**Mg ha^{-1}**		**cm**
1	MF	11.4^{a}	2.1^{d}	52^{b}
1	OG	11.2^{ab}	1.8^{e}	61^{a}
1	PRG-F	9.8^{ab}	2.4^{abc}	38^{cde}
1	PRG-R	10.2^{ab}	2.3^{bcd}	43^{c}
1	PRG-S	9.5^{b}	2.4^{abc}	35^{de}
1	TF	10.2^{ab}	2.2^{cd}	52^{b}
2	MF	7.2^{c}	2.5^{ab}	51^{b}
2	OG	7.3^{c}	2.3^{bcd}	54^{b}
2	PRG-F	4.4^{d}	2.6^{a}	32^{ef}
2	PRG-R	5.0^{d}	2.6^{a}	34^{def}
2	PRG-S	4.2^{d}	2.6^{a}	29^{f}
2	TF	7.0^{c}	2.4^{abc}	39^{cd}
SE		0.5	0.1	1.00
Pr > F		0.0402	0.012	0.0002

$^{a\text{-}f}$ Within each experiment, means without a common superscript differ ($P \leq 0.05$)
*Meadow Fescue (MF); orchardgrass (OG); PRG-F, PRG-R, PRG-S (perennial ryegrass 'Forageur', 'Remington', 'Spreader IV', respectively)

Figure 2. Persistence (% ground cover) of cool-season grasses during the spring following the last grazing event in St. Paul and Grand Rapids, MN.

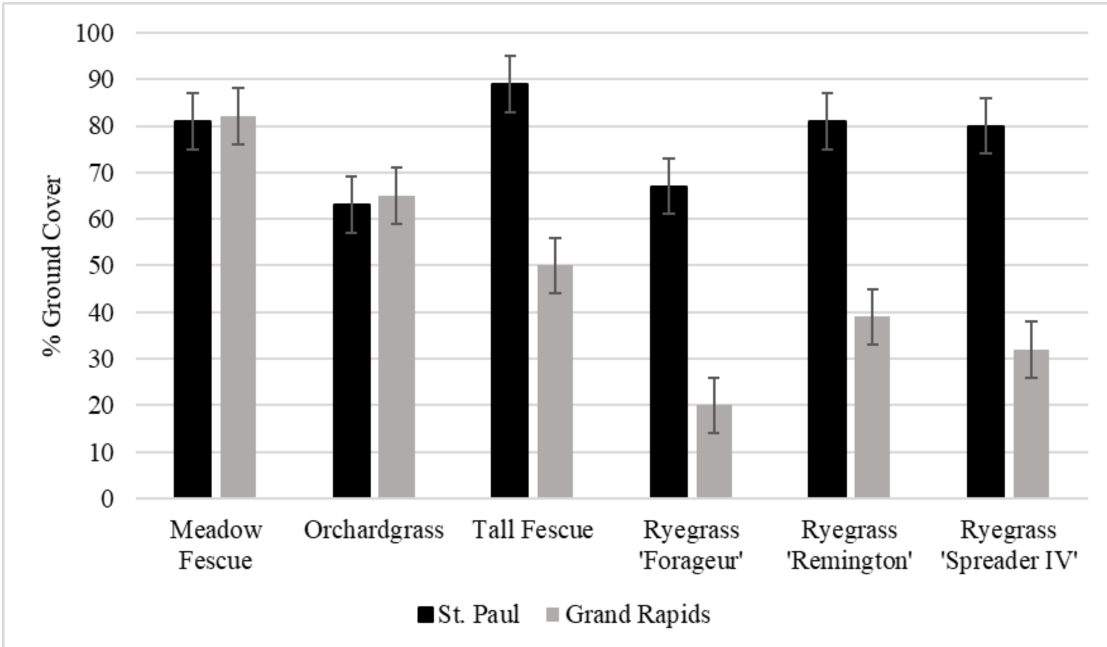

Table 3. Ratio of cool-season grasses and white clover (G:C) under livestock grazing in St. Paul (SP) and Grand Rapids (GR), MN.

Location by Year		G:C
Location	Year	
SP	1	8:1[b]
SP	2	11:1[b]
GR	1	139:1[a]
GR	2	25:1[b]
SE		18
Pr > F		0.0008

[a-b] Within each experiment, means without a common superscript differ ($P \leq 0.05$)

Table 4. Forage nutritive values of cool-season grasses in St. Paul and Grand Rapids, MN under livestock grazing.

Year by Grass		CP	NDF	Ca	P	NSC	Equine DE	Cattle ME	NDFD 48
year	Grass*	g kg^{-1} DM					Mcal/kg		g kg^{-1} NDF
1	MF	185^{def}	576^{bc}	5.16	4.04^{e}	122de	2.25^{bcd}	2.42^{c}	857^{d}
1	OG	184^{def}	601^{a}	3.78	4.19^{bcde}	101^{f}	2.17^{cd}	2.41^{c}	862^{cd}
1	PRG-F	191^{cdef}	537de	4.51	4.16^{cde}	128^{cd}	2.31^{b}	2.47^{abc}	852^{d}
1	PRG-R	192^{bcde}	544de	4.00	4.45^{abc}	132^{bcd}	2.27^{b}	2.54^{a}	886^{bc}
1	PRG-S	205^{bc}	537de	4.65	4.11^{de}	117^{def}	2.31^{b}	2.52^{ab}	916^{a}
1	TF	196^{bcd}	557^{cd}	4.33	4.22^{abcde}	121de	2.27^{bc}	2.48^{abc}	890^{ab}
2	MF	174^{f}	546de	3.91	4.1^{de}	147^{ab}	2.28^{b}	2.44^{bc}	799^{e}
2	OG	176^{ef}	591^{ab}	3.49	4.53^{a}	105^{ef}	2.16^{d}	2.32^{d}	823^{e}
2	PRG-F	209^{ab}	499^{f}	5.11	4.51^{a}	143^{bc}	2.41^{a}	2.44^{bc}	851^{d}
2	PRG-R	198^{bcd}	501^{f}	4.30	4.50^{ab}	162^{a}	2.42^{a}	2.44^{bc}	851^{d}
2	PRG-S	224^{a}	473^{g}	5.41	4.51^{a}	147^{ab}	2.46^{a}	2.51^{ab}	852^{d}
2	TF	197^{bcd}	528^{e}	4.68	4.37^{abcd}	134^{bcd}	2.30^{b}	2.42^{c}	864^{cd}
SE		4.7	7.2	0.04	0.08	4.2	0.02	0.02	7.0
Pr > F		<0.0001	<0.0001	0.0602	0.0154	0.0015	<0.0001	0.0025	<0.0001

[a-f] Within each experiment, means without a common superscript differ ($P \leq 0.05$)
*Meadow Fescue (MF); orchardgrass (OG); PRG-F, PRG-R, PRG-S (perennial ryegrass ‘Forageur’, ‘Remington’, ‘Spreader IV’, respectively)

Table 5. Horse and beef cow preference when grazing cool-season grasses in MN.

Year by Grass		Horse	Beef Cow
Year	Grass	% removal	
1	MF	99[a]	64[ef]
1	OG	84[b]	73[c]
1	PRG-F	99[a]	82[b]
1	PRG-R	98[a]	88[a]
1	PRG-S	98[a]	86[ab]
1	TF	99[a]	66[de]
2	MF	58[e]	68[de]
2	OG	67[d]	62[f]
2	PRG-F	74[c]	70[cd]
2	PRG-R	82[b]	62[f]
2	PRG-S	81[b]	70[cd]
2	TF	70[cd]	70[cd]
SE		4	4
Pr > F		0.0001	0.0001

[a-f] Within each experiment, means without a common superscript differ ($P \leq 0.05$)
*Meadow Fescue (MF); orchardgrass (OG); PRG-F, PRG-R, PRG-S (perennial ryegrass 'Forageur', 'Remington', 'Spreader IV', respectively)

Figure 3. Morphometric measurements collected on Miniature horses, saddle-type horses, and Thoroughbreds, including neck circumference located halfway between the poll and the withers (A), height at the third thoracic vertebra (B), girth circumference at the third thoracic vertebra (C), body length (BL straight) from the point of the shoulder to a point perpendicular to the point of the buttock (D), and body length (BL wrap) from the point of the shoulder to the point of the buttock (E).

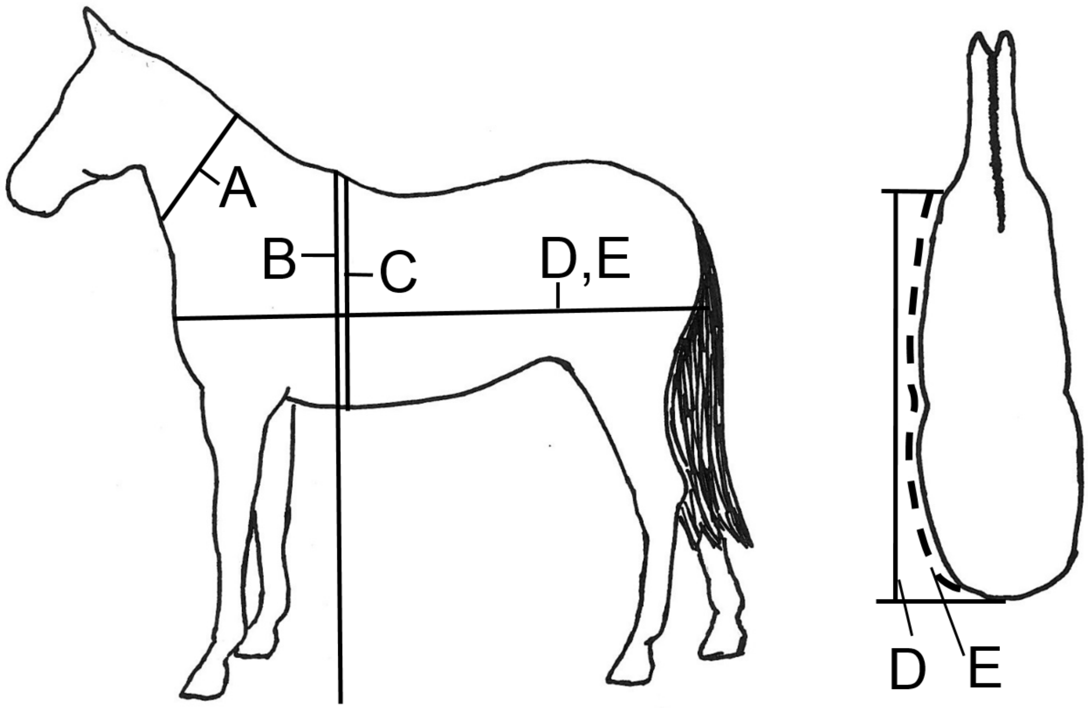

Table 6. Description of breed types, breeds and mean (mean ± SD) age, body condition score (BCS), morphometric measurements and bodyweights of 637 equines obtained from 10 shows or farms from 2014 to 2017.

Age Group	Breed or Breed Registry	Total	BCS = 5	BCS	Age	Height	BL[1] Straight	BL[2] Wrap	Girth. Circ.[3]	Neck Circ.[4]	Scale weight
		n			year			cm			kg
(≥ 3 yr)	Miniature	261	70	6.1 ± 0.8	7.3 ± 4.0	92 ± 7	97 ± 7	97 ± 7	114 ± 8	64 ± 5	117 ± 21
(< 3 yr)	Miniature	62	39	5.4 ± 0.6	1.1 ± 0.6	88 ± 8	90 ± 9	90 ± 9	102 ± 12	56 ± 8	91 ± 27
(≥ 3 yr)	TB[5]	100	68	5.0 ± 0.6	10.2 ± 4.9	164 ± 4	172 ± 7	172 ± 7	192 ± 5	97 ± 5	538 ± 42
(≥ 3 yr)	ASB[6] and crosses	53	16	6.0 ± 1.0	10.5 ± 4.4	157 ± 6	162 ± 7	162 ± 7	186 ± 6	98 ± 6	510 ± 42
	Morgan	48	1	6.6 ± 0.9	9.9 ± 5.5	149 ± 4	161 ± 7	161 ± 7	182 ± 5	97 ± 6	480 ± 31
	Tennessee Walking Horse	88	27	5.7 ± 1.0	11.3 ± 6.0	155 ± 6	155 ± 8	155 ± 8	181 ± 8	85 ± 6	456 ± 56
	Other[7]	25	7	5.6 ± 1.2	11.3 ± 5.2	152 ± 7	152 ± 7	152 ± 7	179 ± 9	86 ± 7	445 ± 63
	Saddle-type mean	214	51	6.0 ± 1.0	10.8 ± 5.3	153 ± 6	158 ± 7	158 ± 7	182 ± 4	92 ± 6	473 ± 48

[1]Body length straight; [2]body length wrap; [3]girth circumference; [4]neck circumference; [5]Thoroughbred; [6]American Saddlebred; and [7]Other owner-reported breeds included Gaited Paint Horse (n=2), Kentucky Mountain Gaited Horse (n=1), Missouri Fox Trotter (n=3), Paso Fino (n=1), Racking Horse (n=5), Rocky Mountain Horse (n=1), Saddle Horse (n=3), Spotted Saddle Horse (n=5) and gaited cross (n=4).

Table 7. Scale bodyweight (BW), predicted ideal bodyweight, and the difference (Diff) between the two based on body condition score (BCS) for adult (≥ 3 years) and juvenile (< 3 years) Miniature, saddle-type and Thoroughbred horses obtained from 10 shows or farms between 2014 and 2017

	Miniature (≥ 3 years)			Miniature (< 3 years)			Saddle			Thoroughbred		
BCS	Scale BW	Ideal BW[a]	Diff	Scale BW	Ideal BW[a]	Diff	Scale BW	Ideal BW[a]	Diff	Scale BW	Ideal BW[a]	Diff
	kg											
3	.[b]	.	.	.	.	.	.	.	.	485±27	548±3	-63±30
4	.	.	.	.	.	.	415±51	444±40	-29±31	523±42	538±23	-15±29
5	110±22	110±21	0±9	84±25	84±25	0±8	451±49	460±39	-9±36	538±37	538±26	0±28
6	117±18	112±19	5±9	101±23	98±23	3±8	476±49	476±41	0±32	564±49	539±28	25±35
7	121±19	112±20	9±9	127±28	123±25	4±3	495±47	484±44	11±31	.	.	.
8	122±20	103±18	19±6	.	.	.	509±43	482±28	27±31	.	.	.

[a]Adult Miniature horse: $[0.00115 \times \text{Length}^{0.786} \times \text{Height}^{1.741}]$, Juvenile Miniature horse: $[0.00005492 \times \text{Length}^{1.919} \times \text{Height}^{1.261}]$, Saddle-type: $[0.00279 \times \text{Length}^{1.370} \times \text{Height}^{1.014}]$, Thoroughbred: $[((1.913 \times \text{Length}) + (4.251 \times \text{Height})) - 487.900]$

[b] No adult Miniature horse received a BCS of 3 or 4 while no juvenile Miniature horse received a BCS of 3, 4 or 8. No saddle-type horse received a BCS of 3, while no Thoroughbred received a BCS of 7 or 8.

Figure 4. Average monthly air and stock tank water temperatures from June through October 2017 in St. Paul, MN.

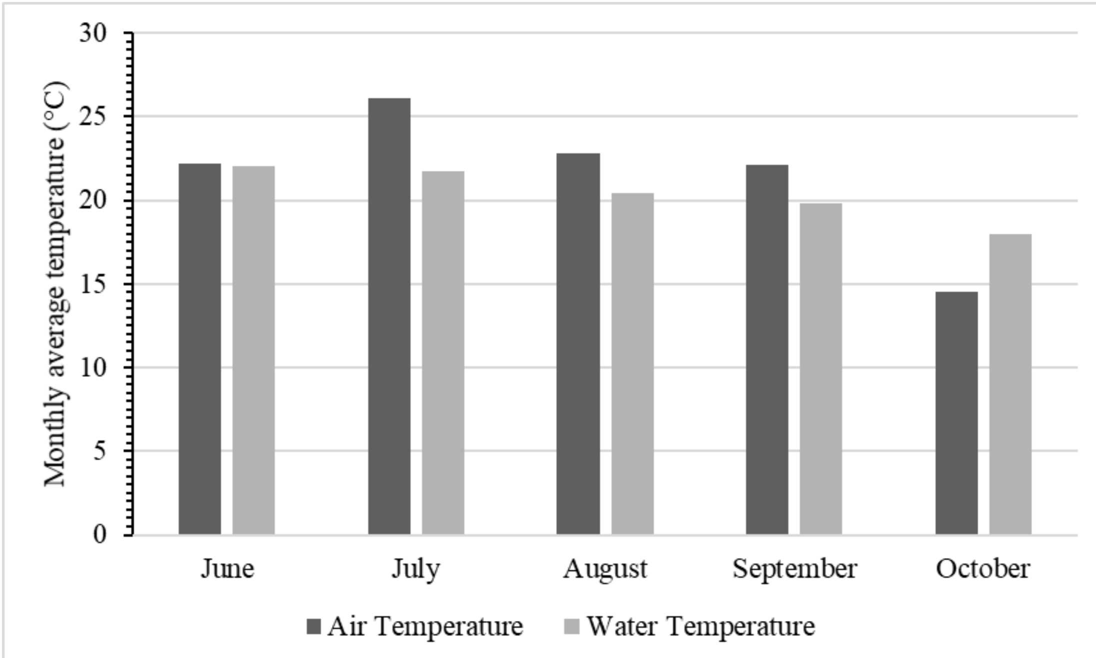

Table 8. Mean water quality parameters, averaged over periods, of plastic and metal water tanks with and without goldfish.

Tank-Type	Inclusion of Goldfish	Total Dissolved Solids (mg/L)	Water Turbidity* (NTU)	Chlorophyll *a* * (mg/m^3)
Plastic	Yes	233^{d}	18.4^{a}	13.2^{a}
	No	235^{c}		
Metal	Yes	254^{b}	9.3^{b}	7.1^{b}
	No	259^{a}		
	SE	3.4	1.7	2.7
	P Value	0.007	< 0.001	0.018

* There was no effect of goldfish ($P \geq 0.11$) on water turbidity or chlorophyll *a*; therefore, values are presented by tank type only.
$^{a-d}$ Within each column, means without a common superscript differ ($P \leq 0.05$).

Table 9. Fish usage in horse water tanks by geographic location.

Location	Currently use fish		Have used fish		Have never used fish		Total
	n	%	n	%	n	%	n
Mid-Atlantic	5	10	3	6	41	84	49
Midwest	74	22	105	32	153	46	332
New England	0	0	5	12	36	88	41
Rocky Mountains	3	17	4	22	12	61	19
South	15	17	24	27	50	56	89
Southwest	6	43	3	21	5	36	14
West	11	28	12	31	17	41	40
Outside U.S.	8	9	18	20	62	71	88
Total (n)	122	-	174	-	376	-	672